

**A FINAL REPORT TO THE
CALIFORNIA DRIED PLUM BOARD**

**EFFECTS OF DRIED PLUMS ON SUPPRESSION OF
GROWTH OF FOODBORNE PATHOGENS IN LIQUID
MEDIUM AND GROUND MEAT**

EXECUTIVE SUMMARY

From

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ABSTRACT

The antimicrobial effect of dried plum in liquid, powder and puree forms was examined in this experiment. The dried plum mixtures tested were United States Department of Agriculture (USDA) dried plum puree in liquid medium (0, 1, 2.5, 3, 5, 7.5, 10% w/v), uncooked ground beef (0, 3% w/w), uncooked pork sausage (0, 3, 6% w/w), and cooked pork sausage (0, 3, 6% w/w); fresh plum juice concentrate in liquid medium (0%, 1%, 2.5%, 5% w/w) and in uncooked ground beef (0, 3% w/w); and Lighter Bake (LB) powder in liquid medium (0%, 1%, 2.5%, 3%, 5% w/w); in uncooked pork sausage (0%, 3%, 6% w/w), and cooked pork sausage (0%, 3%, 6% w/w). The three plum mixtures were obtained from the California Dried Plum Board. Each test mixture was inoculated with a 5-strain pathogen cocktail. Microbial analysis was performed at day 0, 1, 3, and 5.

When compared to the control, the liquid medium with greater than 2.5% dried plum puree, plum juice, or Lighter Bake (LB) powder exhibited a 2 log or greater suppression of total aerobic count, *Salmonella typhimurium*, *Listeria monocytogenes*, *Escherichia coli* O157:H7, *Yersinia enterocolitica*, and *Staphylococcus aureus*. The uncooked ground beef (20% fat) with dried plum puree or plum juice exhibited a 1 log to 2 log suppression of total count, *S. typhimurium*, *L. monocytogenes*, *E. coli* O157:H7, *Y. enterocolitica*, and *S. aureus* when compared to the control after 5 days. The higher fat uncooked pork (29.26-31.26% fat) with 6% LB powder or 6% dried plum puree resulted in a greater than 0.5 log suppression of total aerobic count, *L. monocytogenes*, *E. coli* O157:H7, and *Y. enterocolitica* compared to the control at 5 days. With *S. typhimurium* and *S. aureus*, no significant difference was noted between the control and treatments with dried plum puree and LB powder. The cooked pork sausage (29.26-31.26% fat) with LB powder or dried plum puree resulted in a 1 log suppression of total count with 3% dried plum puree, >1 log suppression of *E. coli* O157:H7 with 6% LB powder and 6% dried plum puree, > 0.5 log suppression with 3% and 6% dried plum puree of *L. monocytogenes*, and >2 log suppression of *Y. enterocolitica* with 3% and 6% dried plum puree compared to the control at 5 days. Because of the effects of pre-cooking, no significant difference was found between the control and the treatments 3% and 6% LB

powder, 3% and 6% dried plum puree, or BHA/BHT for *S. typhimurium* in cooked pork sausage at 5 days.

INTRODUCTION

Salmonella typhimurium, *Listeria monocytogenes*, *Escherichia coli* O157:H7, *Yersinia enterocolitica*, and *Staphylococcus aureus* have been identified as agents of foodborne diseases. Physical, chemical, and biological methods have been used to control them in foods. One approach for the control and prevention of foodborne pathogens is using naturally occurring food ingredients. Spice plants' secondary components are powerful antimicrobials (Billing and Sherman, 1998). At Kansas State University, garlic and cinnamon have reduced *E. coli* O157:H7 in liquid media and ground beef (Ceylan et al., 1999a; 1999b). Previous studies at Kansas State and The Pennsylvania State University have provided information on the killing effects of phenolic antioxidants, such as BHA, BHT, TBHQ, and PG, on pathogenic fungi and numerous bacteria including foodborne pathogens (Fung et al., 1985; Fung et al., 1977; Gailani and Fung, 1984; Lin and Fung, 1983).

Commercial dried plum and dried plum extracts (*Prunus domestica* cv. French) contain phenolics, such as hydroxycinnamates, neochlorogenic acid and chlorogenic acid, which can inhibit the oxidation of low-density lipoprotein (Nakatani et al, 2000). Kreuzer (2001) reported that 3% dried plum puree worked comparably to BHA/BHT on prevention of warmed-over flavor caused by lipid oxidation in precooked pork sausage. The effect of dried plum extracts on foodborne pathogens has not been reported.

The objective of this study was to evaluate the efficacy of different concentrations and various times of contact of dried plum mixtures for controlling *S. typhimurium*, *L. monocytogenes*, *E. coli* O157:H7, *Y. enterocolitica*, and *S. aureus* in liquid medium, uncooked ground beef, uncooked pork sausage, and cooked pork sausage.

MATERIALS AND METHODS

Inoculum Preparation

The inoculum for this study consisted of 1 strain of each of 5 different organisms. The organisms were *Yersinia enterocolitica* 0:8, *Salmonella typhimurium*, *Escherichia coli* O157:H7 ATCC 35150, *Listeria enterocolitica* Scott A ATCC 49594, and *Staphylococcus aureus* from the culture collection of Kansas State University Food Microbiology Lab. The cultures were tested for purity using Gram reactions and biochemical tests: Enterotube (BBL, Becton Dickinson, Sparks, MD), API 20E (bioMerieux, Inc., Hazelwood, MO), Gram-positive ID (BBL, Becton Dickinson, Sparks, MD), and api Listeria (bioMerieux, Inc., Hazelwood, MO).

The five organism cocktail was prepared by growing each organism separately in 100 ml of Brain Heart Infusion (BHI) broth and incubated at 35°C for 24 h. The cultures were then centrifuged at 15,300 x g for 20 minutes, the supernatant was removed and the pellets were resuspended with 100 ml of 0.1% sterile peptone water. The re-suspended cultures were then combined in equal amounts in a sterile glass bottle prior to use. This tube contains ca. 7 log CFU/ml. Target concentration in liquid medium and solid foods was 4 log CFU/ml or g respectively.

Table 1: Methods for Enumerating Inoculated Pathogens

<u>Pathogen Enumerated</u>	<u>Media and Source</u>	<u>Colony Reaction*</u>
<i>Escherichia coli</i> O157:H7	MacConkey Sorbitol Agar ^a (MSA)	Pink or opaque
<i>Staphylococcus aureus</i>	Baird-Parker Agar ^a (BP)	Black w/ clear zone
<i>Listeria monocytogenes</i>	Modified Oxford Medium ^a (MOX)	White w/ black zone
<i>Salmonella typhimurium</i>	Xylose Lysine Deoxycholate ^a (XLD)	Black
<i>Yersinia enterocolitica</i>	Yersinia Selective Agar ^b (CIN)	Pink

* Experimentally determined

^a Source is DIFCO LABORATORIES, Detroit, MI.

^bSource is Oxoid, LTD., Basingstoke, Hampshire, England

Dried Plum Mixtures

All the dried plum mixtures used in these experiments were obtained from the California Dried Plum Board and manufactured by Sunsweet Growers of Yuba City.

Pork Sausage

The pork sausage for this experiment was prepared at Texas A&M University and supplied to the project through the courtesy of Dr. Jim Keeton. Fresh, well-chilled coarse ground (1.27 cm plate) lean and fat pork trimmings were ground separately, combined in a mixer/blender to the appropriate target fat level and reground through a (0.4763 cm or 3/16 inch plate) prior to stuffing into 2 inch plastic casing.

The fresh pork sausage (32-34% fat) was manufactured to contain 0.02% (based on fat content) BHA/BHT combination, or one of two levels (3% and 6%) of dried plum puree and light bake powder. The sausage also contained 3.0% water, 1.5% salt (~2.5% cooked), ~2% seasonings (black pepper, red pepper, sage, sugar). The 1 lb. chubs were then frozen and the product to be tested was shipped to Kansas State University.

Preparation of Sample Materials

Liquid Medium

The liquid medium was prepared by adding 0, 1, 2.5, 5, 7.5, or 10% wt/vol of dried plum mixture to one of 6 sterile bottles containing 100 ml, 99 ml, 97.5 ml, 95 ml, 92.5 ml, and 90 ml of Brain Heart Infusion (BHI) broth respectively and mixed. The media was sterilized prior to addition of mixtures. For light bake powder, a concentration of 0%, 1%, 2.5%, 3%, or 5% was used. A concentration of 0%, 1%, 2.5% or 5% of plum juice concentrate was added to the liquid medium. The bottles were then inoculated with 1 ml of 6 log CFU/ml of the five organisms cocktail. The target initial concentration was 4 log CFU/ml. After the bottles were thoroughly mixed, they were allowed to set at room temperature for 1 hour prior to testing. After sampling, the bottles were placed in an incubator at 35°C until the next sampling day. Each day 1 ml of sample was removed for testing and then the bottle was returned to the incubator.

Uncooked Ground Beef

The ground beef for this experiment was obtained from a retail store the day that the mixtures were made. The mixtures were made by mixing, by sterile gloved hand, 3.75 g of the product dried plum puree or plum juice concentrate with 121.5 g of ground beef (20% fat) until well blended to make mixtures with 3% dried plum puree or plum juice concentrate. The fat percentage of the meat mixtures will decrease slightly as the dried plum mixtures are added. The meat blends and ground beef with no addition of dried plum mixture were then inoculated by adding 1.25 ml of cocktail at 5 log CFU/ml and then mixing for 5 minutes with sterile-gloved hands. The target initial concentration of pathogen was 3 log CFU/g. The inoculated meat was then allowed to rest for 1 hr prior to further manipulation. Each inoculated meat mixture was then divided into 25 g samples with each placed into a sterile filter stomacher bag (Spiral Biotech, model SFB-410, Bethesda, MD) for further testing or for storage. The samples were stored at 4°C until sampling.

Uncooked Pork Sausage

The uncooked pork sausage (34% fat with less in mixtures containing 0% dried plum mixtures) was manufactured at Texas A & M University, frozen and then shipped to our laboratory. After arrival from Texas A&M University, the product was kept frozen until use. Before sampling, the product was allowed to thaw overnight at 4°C. This product was inoculated and separated using the same procedure as was used with the uncooked ground beef.

Cooked Pork Sausage

The uncooked pork sausage prepared by Texas A&M University, were removed from the freezer and placed at 4°C for 24 hours prior to cooking. The chubs were then crust frozen at -40°C for 15 minutes to aid in slicing. Two-inch slices were then taken of each product and placed on a baking sheet. The slices were then placed at 4°C for 20 minutes to allow for temperature equalization. The slices were then placed in a convection oven set at 300°F to cook for 8 minutes to 160°F internal temperature. The slices were then allowed to cool to room temperature. The uninoculated patties were quartered and each 1/4th was weighed and placed into a stomacher bag. The patties to be inoculated were then weighed

individually to determine the amount of inoculum to be added. The 5 log CFU/ml cocktail was added to the patties using a syringe to inject 10 spots (0.1 ml per spot) for a final inoculum level of 3 log CFU/g. These were allowed to rest for 1 hour prior to being sliced into quarters with each 1/4th being weighed and placed into a separate stomacher bag. The samples were stored at 4°C until sampling.

Sampling Methods

Liquid Medium

After the dried plum and Brain Heart Infusion (BHI) broth mixtures were allowed to rest for 1 hour, 1 ml was diluted, using 1 ml of sample into 9 ml of 0.1% peptone water, to detectable levels and duplicate spread plated onto Tryptic Soy Agar (TSA), MacConkey Sorbitol Agar (MSA) for isolating and differentiating enteropathogenic *Escherichia coli* serotypes, Modified Oxford Medium (MOX) for isolation and differentiating *Listeria monocytogenes*, Yersinia Selective Agar (CIN) for isolation and differentiating *Yersinia enterocolitica*, Baird-Parker Agar (BP) for isolation and differentiating *Staphylococcus*, and Xylose Lysine Deoxycholate (XLD) for isolation and differentiating gram-negative bacilli in this case *Salmonella typhimurium*. The BHI broth mixtures were then returned to the incubator at 35°C until the next sampling period. The plates were inverted and incubated at 35°C for 24 hours. After incubation, the plates were counted for typical colonies and recorded as CFU/ml. The same procedure was repeated for sampling after 1, 3 and 5 days. Bacterial counts from BHI with dried plum mixtures were compared to BHI with no additives on the same day and between days. Three repetitions of the experiment were performed.

Uncooked Product

On days 0, 1, 3, and 5, the sample had 225 ml of 0.1% peptone water added to the 25g meat sample in the stomacher bag to make a 1:10 dilution. The contents were then stomached (Lab Blender Stomacher 400, model BA 612, A.J. Seward, London) for 2 minutes and diluted as previously described. The samples were then spiral plated in duplicate onto TSA, MSA, MOX, CIN, BP, and XLD using a Spiral Plater (Spiral Biotech, model D, Bethesda, MD). The plates were inverted and incubated at 35°C for 24 hours. After incubation, the plates were counted and recorded. Three repetitions of the experiment were performed.

Cooked Product

On days 0, 1, 3, and 5 samples were diluted using 0.1% peptone water. On day 0, the samples were diluted at 1:2.5. On the remaining days the samples were diluted at 1:3.5. The samples were then stomached and diluted as described previously. The samples were then spiral plated onto TSA, MSA, MOX, CIN, BP, and XLD using a Spiral Plater. The plates were inverted and incubated at 35°C for 24 hours. After incubation, the plates were counted and recorded. Three repetitions of the experiment were made.

Statistical Analysis

All data presented are averages of three replications and the numbers are statistically analyzed. Statistical analysis of microbial data was performed using Proc GLM Analysis (General Linear Model) and least square means analysis (LSM). Logarithmic transformation of natural numbers was made for statistical analysis. The level of significance among mean values was determined at the 5% level ($\alpha=0.05$).

RESULTS AND DISCUSSION

Dried Plum Mixtures in Liquid Medium

Dried Plum Puree

A summarization of the effect of dried plum puree concentration and contact time in liquid medium by treatment is found in Table 2. Analysis of treatments compared to the control was performed on four days (Day 0,1,3, and 5).

For dried plum puree, the greatest suppression of the total aerobic count, *S. typhimurium*, and *E. coli* O157:H7 and *L. monocytogenes* by dried plum puree is seen on day 5 with 10%. The greatest suppression of *Y. enterocolitica* was on day five with 2.5, 5, and 7.5%. The greatest suppression of *S. aureus* was seen on day 3 with 10%. In general, at 2.5 to 10% there was 2 log CFU/ml or greater suppression of pathogens by dried plum puree compared with the control at 5 days.

Table 2: Dried Plum Puree in Liquid Medium

Treatment	Day	Total Count Log CFU/ml	<i>S. typhimurium</i> Log CFU/ml	<i>E. coli</i> O157:H7 Log CFU/ml	<i>L.monocytogens</i> Log CFU/ml	<i>Y. enterocolitica</i> Log CFU/ml	<i>S. aureus</i> Log CFU/ml
Control	0	4.57 ^{ab}	3.81 ^b	3.75 ^b	3.63 ^{ab}	3.07 ^b	3.78 ^{ab}
	1	9.35 ^{ef}	8.99 ^{ef}	8.59 ^{fg}	7.58 ^e	6.48 ^{cd}	7.11 ^{bc}
	3	8.68 ^{def}	8.11 ^{ef}	7.77 ^{ef}	7.24 ^{de}	5.61 ^{cd}	6.04 ^{bc}
	5	8.31 ^d	7.84 ^{de}	6.19 ^{cd}	6.86 ^{de}	4.95 ^c	4.46 ^{ab}
1%	0	4.39 ^{ab}	3.66 ^b	3.69 ^b	3.54 ^{ab}	3.22 ^{bc}	3.72 ^{ab}
	1	9.22 ^{ef}	8.54 ^{ef}	8.87 ^{fg}	6.86 ^{de}	6.95 ^d	6.87 ^{bc}
	3	8.71 ^{def}	7.96 ^{de}	8.19 ^{efg}	6.64 ^{de}	4.11 ^{bc}	5.38 ^{bc}
	5	8.03 ^d	7.5 ^{de}	6.94 ^{de}	5.4 ^{cd}	2.55 ^b	3.26 ^a
2.5%	0	4.87 ^{ab}	3.66 ^b	3.76 ^b	3.59 ^{ab}	3.44 ^{bc}	3.7 ^{ab}
	1	9.16 ^{ef}	8.29 ^{ef}	8.65 ^{fg}	6.31 ^d	5.18 ^c	5.54 ^{bc}
	3	6.7 ^c	5.24 ^c	5.64 ^{cd}	6.08 ^{cd}	2.06 ^{ab}	3.79 ^{ab}
	5	5.77 ^b	4.16 ^b	3.21 ^{ab}	4.69 ^{bc}	0.33 ^a	3.13 ^a
5%	0	4.25 ^a	3.63 ^b	3.74 ^b	3.51 ^{ab}	3.48 ^{bc}	3.78 ^{ab}
	1	8.43 ^{de}	8.14 ^{ef}	7.88 ^{ef}	5.9 ^{cd}	4.98 ^c	5.52 ^{bc}
	3	7.07 ^c	4.54 ^{bc}	5.25 ^c	5.97 ^{cd}	1.67 ^{ab}	3.63 ^{ab}
	5	5.73 ^b	3.96 ^b	3.44 ^b	4.67 ^{bc}	0.33 ^a	2.37 ^a
7.5%	0	4.59 ^{ab}	3.76 ^b	4.32 ^{bc}	3.49 ^{ab}	3.4 ^{bc}	3.87 ^{ab}
	1	8.79 ^{def}	7.96 ^{de}	7.88 ^{ef}	5.56 ^{cd}	5.38 ^{cd}	4.47 ^{ab}
	3	7.13 ^c	5.37 ^c	5.7 ^{cd}	5.5 ^{cd}	3.5 ^{bc}	3.57 ^{ab}
	5	5.52 ^b	3.27 ^{ab}	2.49 ^{ab}	4.41 ^{bc}	0.33 ^a	3.34 ^{ab}
10%	0	4.22 ^a	3.65 ^b	3.8 ^b	3.13 ^a	3.43 ^{bc}	3.27 ^{ab}
	1	8.71 ^{def}	7.07 ^d	7.51 ^e	5.55 ^{cd}	4.91 ^c	4.47 ^{ab}
	3	7.43 ^c	6.09 ^{cd}	6.36 ^d	5.28 ^c	1.67 ^{ab}	3.35 ^{ab}
	5	5.15 ^b	2.41 ^a	2.33 ^a	4.15 ^b	0.67 ^a	2.46 ^a

^{a-f} = Data with the same letter under each organism represents data that is not significantly different from each other ($\alpha > 0.05$)

Lighter Bake Powder

The percentage of dried plum mixtures was decreased after learning that the functional amount to add to not change the taste of the food was 6%. A summary of the effect of LB powder concentration and contact time in liquid medium between treatments is found in Table 3.

For LB powder, the greatest suppression of total aerobic count and *L. monocytogenes* was seen on day 5 with 3%. For *S. typhimurium*, *E. coli* O157:H7, and *S. aureus*, the greatest suppression was seen on day 5 with 5%. For *Y. enterocolitica*, the greatest suppression was seen with 2.5, 3, and 5%. In general, at 2.5% to 5%, there was a 2 log CFU/ml or greater suppression of pathogens by LB powder compared with the control at 5 days.

Table 3: Lighter Bake Powder in Liquid Medium

Treatment	Day	Total Count Log CFU/ml	<i>S.typhimurim</i> Log CFU/ml	<i>E. coli</i> O157:H7 Log CFU/ml	<i>L.monocytogenes</i> Log CFU/ml	<i>Y.enterocolitica</i> Log CFU/ml	<i>S. aureus</i> Log CFU/ml
0	0	5.33 ^{ab}	4.38 ^b	4.61 ^{bc}	4.50 ^{bc}	3.69 ^{cd}	4.62 ^c
	1	9.12 ^d	8.68 ^f	8.92 ^d	7.54 ^{de}	6.33 ^e	5.83 ^d
	3	8.90 ^{cd}	8.66 ^{ef}	8.27 ^d	7.47 ^{de}	6.23 ^e	4.95 ^{cd}
	5	8.41 ^{cd}	8.44 ^{ef}	6.17 ^c	7.38 ^{de}	5.57 ^e	4.30 ^c
1	0	5.22 ^a	4.41 ^b	4.61 ^{bc}	4.39 ^b	3.99 ^{cd}	4.55 ^c
	1	9.07 ^d	8.55 ^{ef}	8.90 ^d	7.15 ^{cde}	6.22 ^e	6.25 ^d
	3	8.65 ^{cd}	6.18 ^{cd}	8.22 ^d	6.00 ^c	2.23 ^b	2.96 ^b
	5	8.17 ^{cd}	5.44 ^c	6.10 ^c	5.11 ^{bc}	1.42 ^b	2.20 ^{ab}
2.5	0	5.38 ^{ab}	4.52 ^{bc}	4.76 ^{bc}	4.32 ^b	4.08 ^{cd}	4.59 ^c
	1	7.80 ^c	6.75 ^d	7.83 ^d	6.15 ^{cd}	4.51 ^d	5.77 ^d
	3	7.30 ^{bc}	5.23 ^{bc}	5.77 ^c	5.82 ^c	2 ^b	2.44 ^{ab}
	5	5.04 ^a	4.67 ^{bc}	4.08 ^b	4.28 ^{ab}	0 ^a	2.04 ^{ab}
3	0	5.46 ^{ab}	4.46 ^b	4.72 ^{bc}	4.53 ^{bc}	4.08 ^{cd}	4.66 ^{cd}
	1	9.01 ^{cd}	7.74 ^e	8.78 ^d	6.94 ^{cde}	3.61 ^c	5.82 ^d
	3	6.52 ^b	4.54 ^{bc}	5.38 ^c	6.11 ^{cd}	1.67 ^b	2.6 ^b
	5	4.87 ^a	3.06 ^a	3.17 ^{ab}	3.79 ^a	0 ^a	2.19 ^{ab}
5	0	5.45 ^{ab}	4.53 ^{bc}	4.83 ^{bc}	4.58 ^{bc}	4.08 ^{cd}	4.79 ^{cd}
	1	8.92 ^{cd}	8.11 ^{ef}	8.75 ^d	6.85 ^{cde}	4.44 ^d	6.18 ^d
	3	6.82 ^{bc}	4.33 ^b	5.64 ^c	6.36 ^{cde}	2 ^b	2.08 ^{ab}
	5	5.2 ^a	2.38 ^a	2.59 ^a	3.96 ^a	0 ^a	1.41 ^a

^{a-e} = Data with the same letter under each organism represents data that is not significantly different from each other ($\alpha > 0.05$)

Plum Juice Concentrate

A summary of the effect of plum juice concentrate concentration and contact time in liquid medium between treatments is found in Table 4

For plum juice concentrate, the greatest suppression of total aerobic count, *S. typhimurium*, *E. coli* O157:H7, and *L. monocytogenes* was seen on day 5 with 5%. *Y. enterocolitica* counts are reduced the most by 2.5% and 5%. In general, at 2.5% to 5%, there was a 2 log CFU/ml or greater suppression of pathogens by plum juice concentrate compared with the control at 5 days.

Table 4: Plum Juice in Liquid Medium

Treatment	Day	Total Count Log CFU/ml	S. typhimurium Log CFU/ml	E. coli O157:H7 Log CFU/ml	L. monocytogenes Log CFU/ml	Y. enterocolitica Log CFU/ml	S. aureus Log CFU/ml
0	0	4.35 ^a	3.14 ^{ab}	3.17 ^a	3.77 ^a	2.95 ^c	3.77 ^b
	1	9.13 ^{de}	7.72 ^c	8.39 ^{de}	7.67 ^c	5.98 ^{de}	4.04 ^b
	3	9.12 ^{de}	7.81 ^c	8.35 ^{de}	7.65 ^c	6.08 ^{de}	3.46 ^{ab}
	5	8.91 ^{de}	7.78 ^c	6.68 ^c	7.65 ^c	6.64 ^e	3.34 ^{ab}
1	0	4.43 ^a	3.32 ^{ab}	3.12 ^a	3.78 ^a	3.09 ^c	3.88 ^b
	1	9.41 ^{de}	7.27 ^c	9.26 ^{de}	7.66 ^c	6.65 ^e	3.47 ^{ab}
	3	8.32 ^{de}	4.31 ^b	7.31 ^{cd}	7.25 ^c	2.33 ^{bc}	3.11 ^{ab}
	5	6.62 ^{bc}	4.38 ^b	5.11 ^b	5.67 ^{bc}	0.26 ^{ab}	2.16 ^a
2.5	0	4.53 ^{ab}	3.7 ^{ab}	3.82 ^{ab}	3.93 ^a	3.38 ^c	4.01 ^b
	1	8.93 ^{cde}	6.99 ^c	8.94 ^{de}	7.05 ^c	5.15 ^d	3.94 ^b
	3	6.87 ^c	3.48 ^{ab}	5.61 ^{bc}	6.62 ^c	1 ^{ab}	2.34 ^{ab}
	5	5.61 ^b	3.17 ^{ab}	3.06 ^a	5.19 ^b	0 ^a	2.13 ^a
5	0	4.62 ^{ab}	3.7 ^{ab}	3.85 ^{ab}	3.85 ^a	3.49 ^{cd}	4.16 ^b
	1	8.85 ^{de}	7.4 ^c	8.84 ^{de}	7.16 ^c	4.18 ^{cd}	3.98 ^b
	3	7.26 ^c	3.47 ^{ab}	6.4 ^{bc}	5.96 ^{bc}	1.33 ^b	2.24 ^a
	5	5.15 ^{ab}	2.55 ^a	2.96 ^a	4.25 ^{ab}	0 ^a	2.64 ^{ab}

a-e = Data with the same letter under each organism represents data that is not significantly different from each other ($\alpha > 0.05$)

Uncooked Ground Beef

The concentration of 3% of dried plum mixtures was used because it provided the same amount of suppression compared to other concentrations while also being functional in meat products for antioxidant capabilities while not effecting the flavor. A summary of significant differences of the effect of dried plum puree and plum juice concentrate and contact time in uncooked ground beef between treatments is found in Table 5.

In general, there was a 1 to 2 log CFU/ml suppression of pathogens with both LB powder and dried plum puree compared with the control at 5 days. The growth of the pathogens at 4°C is unusual but is supported by work reported by Harshavaerhan (1992).

Table 5: Dried Plum Mixtures in Ground Beef (20% Fat)

Treatment	Day	Total Count Log CFU/ml	<i>S.typhimurium</i> Log CFU/ml	<i>E. coli</i> O157:H7 Log CFU/ml	<i>L. monocytogenes</i> Log CFU/ml	<i>Y.enterocolitica</i> Log CFU/ml	<i>S. aureus</i> Log CFU/ml
<i>Control</i>	0	5.06 ^a	4.3 ^{ab}	4.25 ^a	3.8 ^{ab}	3.92 ^a	3.69 ^a
	1	6.56 ^{bc}	5.23 ^c	5.37 ^b	3.85 ^{ab}	5.46 ^b	4.63 ^{bc}
	3	7.14 ^c	4.9 ^{bc}	6.85 ^{cd}	3.71 ^{ab}	6.68 ^c	4.46 ^{bc}
	5	8.79 ^d	5.16 ^{bc}	7.52 ^d	4.21 ^b	6.97 ^c	4.77 ^{bc}
3% Dried Plum Puree	0	5 ^a	4.25 ^{ab}	4.36 ^a	4.05 ^{ab}	3.99 ^a	3.98 ^{abc}
	1	5.8 ^b	4.94 ^{bc}	4.82 ^{ab}	4.02 ^{ab}	4.9 ^{ab}	4.23 ^{abc}
	3	6.54 ^c	4.57 ^{bc}	5.17 ^b	3.6 ^{ab}	5.32 ^b	4.02 ^{abc}
	5	7.3 ^c	4.03 ^a	5.57 ^c	3.52 ^a	5.21 ^{bc}	3.94 ^{abc}
3% Plum Juice	0	5.01 ^a	4.3 ^{ab}	4.18 ^{ab}	3.85 ^{ab}	3.86 ^{ab}	4.2 ^{ab}
	1	6.11 ^{ab}	4.68 ^{bc}	4.66 ^{ab}	3.76 ^{ab}	4.6 ^{ab}	4.36 ^{abc}
	3	7.13 ^{bc}	4.59 ^b	5.48 ^b	3.77 ^{ab}	5.12 ^b	4.37 ^{ab}
	5	7.26 ^c	3.76 ^{ab}	6.5 ^b	3.48 ^a	6.46 ^b	4.14 ^{ab}

^{a-c}= Data with the same letter under each organism represents data that is not significantly different from each other (x>0.05)

Uncooked Pork Sausage

The concentrations of 3% and 6% were chosen to test because they fall within the functional level of 3-6%. The 3% concentration in uncooked ground beef was seen to not have as high a suppression of organisms as seen with liquid medium. We then decided to test at the functional level to see if there was increased suppression of test organisms compared to the control. A summary of the effect of LB powder, dried plum puree and BHT/BHA concentrate and contact time in uncooked pork sausage between treatments is found in Table 6.

Table 6: Dried Plum Mixtures in Uncooked Pork Sausage (34% Fat)

Treatment	Day	Total Count log CFU/g	<i>S.typhimurm</i> log CFU/g	<i>E.coli</i> O157:H7 log CFU/g	<i>L.monocytogens</i> log CFU/g	<i>Y.enterocolitia</i> log CFU/g	<i>S. aureus</i> <i>log CFU/g</i>
Control	0	6.98 ^{bc}	2.1 ^{ab}	5.13 ^{ab}	2.62 ^a	5.22 ^b	2.89 ^{abc}
	1	8.09 ^{cde}	1.98 ^a	6.01 ^b	2.29 ^a	5.99 ^{bc}	3.18 ^{bc}
	3	8.45 ^{de}	1.98 ^a	6.60 ^{bcd}	3.09 ^a	7.41 ^{cd}	3.18 ^{bc}
	5	8.78 ^{de}	2 ^a	7.85 ^{cd}	2.99 ^a	7.92 ^d	3.28 ^{bc}
Lighter Bake 3%	0	6.21 ^{ab}	2.2 ^{ab}	4.88 ^a	2.23 ^a	4.86 ^{ab}	2.68 ^{ab}
	1	6.73 ^b	2.26 ^{ab}	5.61 ^{ab}	2.52 ^a	6.05 ^{bc}	2.85 ^{ab}
	3	7.94 ^{cd}	1.98 ^a	6.84 ^{bcd}	2.37 ^a	6.78 ^c	3.29 ^{bc}
	5	8.32 ^{cde}	1.98 ^a	7.00 ^{bcd}	2.59 ^a	7.11 ^{cd}	3.21 ^{bc}
Lighter Bake 6%	0	6.22 ^{ab}	2.1 ^{ab}	5.08 ^{ab}	2.39 ^a	4.64 ^{ab}	2.69 ^{ab}
	1	6.51 ^b	2.35 ^b	5.43 ^{ab}	2.64 ^a	5.52 ^b	3.17 ^{bc}
	3	7.61 ^{cd}	2.1 ^{ab}	5.75 ^{ab}	2.28 ^a	6.19 ^{bc}	3.01 ^{abc}
	5	7.96 ^{cd}	1.98 ^a	6.32 ^{bc}	2.64 ^a	6.15 ^{bc}	2.87 ^{abc}
Dried Plum Puree 3%	0	6.2 ^{ab}	2.13 ^{ab}	5.10 ^{ab}	2.83 ^a	4.64 ^{ab}	2.64 ^a
	1	7.18 ^{bc}	1.98 ^a	6.25 ^{bc}	2.27 ^a	5.98 ^{bc}	2.90 ^{abc}
	3	8.03 ^{cde}	1.98 ^a	7.00 ^{bcd}	2.18 ^a	6.69 ^c	3.04 ^{abc}
	5	8.40 ^{de}	1.98 ^a	7.21 ^{cd}	2.61 ^a	7.27 ^{cd}	3.10 ^{abc}
Dried Plum Puree 6%	0	5.51 ^a	2.29 ^{ab}	5.01 ^{ab}	2.47 ^a	3.84 ^a	2.65 ^a
	1	6.21 ^{ab}	2.43 ^b	5.11 ^{ab}	2.33 ^a	4.44 ^{ab}	2.77 ^{ab}
	3	7.12 ^{bc}	2.18 ^{ab}	5.86 ^{ab}	2.47 ^a	5.56 ^b	3.3 ^b
	5	7.63 ^{cd}	2.33 ^b	5.4 ^{ab}	2.45 ^a	5.28 ^b	3.28 ^{bc}
BHA/BHT	0	5.49 ^a	2.06 ^{ab}	5.24 ^{ab}	2.70 ^a	4.29 ^{ab}	2.67 ^a
	1	6.62 ^b	2.49 ^b	5.3 ^{ab}	3.62 ^{ab}	5.70 ^{bc}	3.19 ^{bc}
	3	7.78 ^{cd}	2.21 ^{ab}	7.01 ^{bcd}	4.26 ^{ab}	5.49 ^b	3.34 ^{bc}
	5	8.39 ^{de}	1.98 ^a	7.43 ^{cd}	4.81 ^b	6.78 ^c	3.28 ^{bc}

^{a-e} = Data with same letter under each organism is not significantly different from each other ($\alpha > 0.05$)

Contrary to laboratory medium studies when pure mixed cultures were studied and counted, in the meat study normal microbial flora also competes with inoculated cultures. The colonies on selective agar plates were more diversified. Every effort was made to count typical colonies on selective agar plates. Occasionally it was difficult to separate target colonies from background flora. In general, at 6%, there was a 0.5 log CFU/ml or greater suppression of total aerobic count, *E. coli* O157:H7, *L. monocytogenes*, and *Y. enterocolitica* by LB powder and dried plum puree compared with the control at 5 days.

Cooked Pork Sausage

A summary of the effect of LB powder, dried plum puree and BHT/BHA concentrate and contact time in cooked pork sausage between treatments is found in Table 7.

On Day 5, there was a significant suppression of total aerobic count with 3% dried plum puree compared to the control. There was a significant suppression of *E. coli* O157:H7 and *Y. enterocolitica* with 3% dried plum puree and 6% dried plum puree compared to the control.

Table 7: Dried Plum Mixtures in Cooked Pork Sausage (34% Fat)

<i>Treatment</i>	<i>Day</i>	<i>Total Count</i> Log CFU/g	<i>S. typhimurim</i> Log CFU/g	<i>E.coli O157H7</i> Log CFU/g	<i>L. monocytogenes</i> Log CFU/g	<i>Y. enterocolitica</i> Log CFU/g	<i>S. aureus</i> Log CFU/g
Control	0	3.42 ^{ab}	2.54 ^{bc}	2.51 ^{cdefgh}	2.42 ^b	2.02 ^{bc}	2.93 ^{abcd}
	1	3.15 ^a	2.23 ^{bc}	2.04 ^{bcdef}	1.54 ^a	1.93 ^{bc}	2.50 ^a
	3	3.34 ^{ab}	2.44 ^{bc}	2.14 ^{bcdefg}	2.55 ^{bc}	2.68 ^{cdefg}	2.94 ^{abcd}
	5	3.68 ^{bcd}	1.33 ^{ab}	2.65 ^{cdefgh}	2.86 ^{bc}	3.5 ^{cdefg}	2.94 ^{abcd}
Lighter Bake Powder 3%	0	3.50 ^b	2.86 ^c	2.27 ^{bcdefgh}	2.44 ^b	2.04 ^{bc}	2.93 ^{abcd}
	1	3.50 ^b	2.45 ^{bc}	2.74 ^{cdefgh}	2.58 ^{bc}	2.65 ^{bcdefg}	3.25 ^{bed}
	3	3.73 ^{bcd}	2.89 ^c	2.37 ^{bcdefgh}	2.48 ^b	2.87 ^{cdefg}	3.20 ^{bed}
	5	3.93 ^{cd}	2.28 ^{bc}	1.85 ^{bcd}	2.88 ^{bc}	3.67 ^{cdefg}	3.23 ^{bed}
Lighter Bake Powder 6%	0	3.55 ^{bc}	2.38 ^{bc}	2.58 ^{cdefgh}	2.49 ^b	2.16 ^{bed}	3.11 ^{bed}
	1	3.59 ^{bed}	2.74 ^c	2.91 ^{cdefgh}	2.59 ^{bc}	1.95 ^{bc}	3.30 ^{bed}
	3	3.53 ^{bc}	2.37 ^{bc}	2.31 ^{bcdefgh}	2.61 ^{bc}	2.50 ^{bcdef}	3.06 ^{bed}
	5	3.80 ^{bcd}	0.89 ^a	1.64 ^b	2.85 ^{bc}	3.37 ^{cdefg}	3.23 ^{bed}
Dried Plum Puree 3%	0	3.62 ^{bcd}	2.80 ^c	2.97 ^{cdefgh}	2.60 ^{bc}	2.34 ^{bcde}	3.07 ^{bed}
	1	3.44 ^{ab}	2.94 ^c	2.37 ^{bcdefgh}	2.64 ^{bc}	2.37 ^{bcde}	3.20 ^{bed}
	3	3.63 ^{bcd}	2.23 ^{bc}	1.90 ^{bcde}	2.41 ^b	3.25 ^{cdefg}	3.22 ^{bed}
	5	3.14 ^a	1.85 ^b	1.70 ^{bc}	2.36 ^b	2.31 ^{bcde}	2.81 ^{bed}
Dried Plum Puree 6%	0	3.19 ^{ab}	2.68 ^{bc}	2.38 ^{bcdefgh}	2.34 ^b	0 ^a	2.90 ^{bed}
	1	3.54 ^{bc}	2.23 ^{bc}	2.81 ^{cdefgh}	2.54 ^{bc}	1.92 ^{bc}	3.26 ^{abc}
	3	3.63 ^{bcd}	2.25 ^{bc}	2.48 ^{cdefgh}	2.42 ^b	2.81 ^{cdefg}	3.20 ^{abcd}
	5	3.69 ^{bcd}	2.10 ^{bc}	0.67 ^a	2.59 ^{bc}	2.57 ^{bcdefg}	2.98 ^{bed}
BHA/BHT	0	3.51 ^b	2.85 ^c	3.05 ^{cdefgh}	2.67 ^{bc}	2.08 ^{bc}	2.63 ^{ab}
	1	3.36 ^{ab}	2.45 ^{bc}	2.54 ^{cdefgh}	2.54 ^{bc}	1.56 ^b	2.87 ^{abcd}
	3	3.41 ^{ab}	2.70 ^c	1.97 ^{bcdef}	2.34 ^b	2.81 ^{cdefg}	2.90 ^{abcd}
	5	3.86 ^{cd}	2.54 ^{bc}	1.41 ^{ab}	3.11 ^c	3.70 ^{cdefg}	3.10 ^{bed}

a-h = Data with the same letter under each organism represents data that is not significantly different from each other ($\alpha > 0.05$)

OVERALL IMPORTANCE OF THE STUDY

The antagonistic effect of certain food components such as fats and protein in food products must be overcome as they may bind and/or solubilize phenolic antioxidants (Raccach, 1984). Gailani and Fung (1984) found that BHA, BHT, TBHQ, and PG were effective in inhibiting bacterial growth of psychrotrophs, coliforms and fecal coliforms in laboratory media. The antioxidant's antimicrobial activities were greatly reduced in ground pork. The hydrophobic phenolic antioxidants, especially the lipid soluble ones, will localize in the lipid portion of the food, reducing their availability for antimicrobial activity. For this reason, increased concentrations are needed in food products to have similar results as seen with liquid medium. Ground beef and pork sausage differ as the fat content is higher in pork sausage.

Results with liquid medium and uncooked meat products may differ as the liquid medium was sterile before the inoculation of the medium with the foodborne pathogen cocktail. The uncooked meat products have a large number of normal flora before the inoculation with the foodborne pathogen cocktail. The normal flora will compete with the inoculated pathogens as well as increasing the microbial load. This can explain decreases in pathogen concentrations seen in control with some pathogens tested.

The addition of dried plum mixtures can control foodborne pathogens in uncooked meat products. All inoculated pathogens in ground beef decreased by 1-2 log CFU/g and decreased in total aerobic count, *E. coli* O157:H7, *L. monocytogenes*, *Y. enterocolitica*, and *S. aureus* of at least 0.5 log CFU/g in uncooked pork sausage.

In cooked meat products, if the product is heated to the correct internal temperature, pathogens will not be viable. Any recontamination of meat products can be further controlled by the preservative effect of these dried plum mixtures. There was 0.5 log CFU/g suppression of total aerobic count, *E. coli* O157:H7, *L. monocytogenes*, and *Y. enterocolitica* at 5 days in cooked pork sausage.

The results of this study indicate that the food industry should use dried plum mixtures at levels of 6% or less. Dr. Keeton of Texas A&M University has found that 3-6% dried plum puree makes juicier pork sausage patties after re-heating. These products when added to pork sausage can therefore produce palatable patties and potentially suppress pathogen growth at a concentration of 6%. These products can also successfully be applied to meat products such as ground turkey/chicken, ground beef, etc.

CONCLUSIONS

1. In liquid medium:

- a. The higher the concentration of dried plum mixture tested, the greater the suppression of inoculated organisms.
- b. Dried plum puree, light bake powder, and plum juice concentrate were capable of suppressing all of the inoculated organisms.
- c. The suppression of organisms facilitated by the addition of dried plum mixtures in liquid medium shows that the dried plum mixtures can be used to control the growth of foodborne pathogens.

2. In uncooked meat:

- d. The addition of dried plum puree, LB powder, and plum juice concentrate is an effective way of suppressing the growth of microorganisms in uncooked ground beef and uncooked pork sausage.
- e. There are other ingredients, e.g. spices and seasonings, in the pork sausage that may effect the suppression of the inoculated pathogens.

3. In cooked pork sausage:

- a. It does appear that cooking to correct internal temperatures may make the use of dried plums less necessary as an antimicrobial agent.

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- b. Cooked meat should be essentially sterile if cooked to the correct internal temperature, if the product is contaminated after cooking the presence of dried plum mixtures in the product can be used to control the foodborne pathogens.
 4. The results of this study indicate that the use of dried plums at 3-6% concentration in meat products will maintain palatability and juiciness while suppressing pathogens and possibly extending shelf-life.
 5. At 3-6% concentration, there is good suppression of microorganisms while being optimum for functionality.
 6. The addition of dried plum mixtures is a strategy that could be very valuable to the food industry because of dried plum's multiple functionalities.

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