

SURVEY OF MICROBIOLOGICAL CONTENT OF
COMMERCIAL BEEF JERKY

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TABLE OF CONTENTS

| Chapter | Page |
|--|------|
| I. INTRODUCTION..... | 1 |
| II. REVIEW OF LITERATURE | |
| Microbiological safety of jerky..... | 3 |
| Compliance guideline for meat and poultry jerky produced by small and very small plants | 3 |
| Antimicrobial treatments used to minimize pathogens during beef jerky production | 6 |
| Using a home-style dehydrator, Holley RA in 19851..... | 6 |
| The rate of inactivation of <i>E. coli</i> O157:H7, Ryu et al., 1999..... | 7 |
| Four methods evaluated by Harrison et al., 2001 | 7 |
| Survival of inoculated <i>E. coli</i> O157:H7, by Albright et al., 2003 | 8 |
| Raisin extracts and raisin puree as antimicrobial by Bower CK et al., 2003..... | 19 |
| Fate of <i>Staphylococcus aureus</i> by Ingham SC et al., 2005..... | 10 |
| Comparison of dehydrator and smokehouse by Harrison et al., 2006..... | 10 |
| Outbreaks of food borne illnesses related to jerky..... | 10 |
| Water activity (Aw)..... | 12 |
| Acidity (pH) | 15 |
| References | 16 |
| III. MICROBIOLOGICAL EVALUATION OF COMMERCIAL BEEF JERKY | |
| Abstract..... | 21 |
| Introduction..... | 22 |
| Materials and Methods..... | 24 |
| Preparation of samples..... | 24 |
| Procedures for microbial analyses | 25 |
| Total aerobic plate count..... | 25 |
| Yeast and Molds | 25 |
| Coliforms | 26 |
| Coagulase positive for staphylococci..... | 26 |
| Test for <i>Escherichia coli</i> O157:H7 | 26 |
| Tests for <i>Salmonella</i> | 28 |
| Acidity (pH) measurement..... | 29 |
| Measurement of water activity..... | 29 |
| Statistical Method..... | 29 |
| Results and Discussion..... | 30 |

| | |
|---|----|
| Conclusion..... | 35 |
| REFERENCES | 36 |
| APPENDIX A..... | 38 |
| <i>Minividas</i> principle and procedure..... | 38 |
| Vitek procedure..... | 40 |
| Buffered peptone water preparation..... | 40 |
| APPENDIX B | 42 |
| Raw data of microbial counts..... | 42 |
| APPENDIX C | 45 |
| Description of jerky samples..... | 45 |
| APPENDIX D | 49 |
| Statistical data | 49 |

LIST OF TABLES

| | Page |
|--|------|
| Table 2-1. Influence of water activity on microbial growth in various foods..... | 14 |
| Table 3-1. Total plate counts of from original flavor jerky..... | 31 |
| Table 3-2. Total plate counts of from teriyaki flavor jerky..... | 31 |
| Table 3-3. Total plate counts of from smoked flavor jerky..... | 32 |
| Table 3-4. Total plate counts of from peppered flavor jerky..... | 32 |
| Table 3-5. Total plate counts of from hot & spicy flavor jerky..... | 32 |
| Table 3-6. Means of total counts of aerobic bacteria by brand..... | 33 |

CHAPTER I

Introduction

Throughout history man has preserved meat by drying. Drying represents one of the oldest methods of food preservation. “Jerky” is a word that comes from the Quechua term “*Charqui*” which means “dried meat” (29). North American Indians made “pemmican” by mixing ground dried meat with dried fruit or suet. African countries use the term “Biltong” for dried meat (32). Drying meat was used in prehistoric times to preserve hunted animals that were too big to eat all at once. Jerky has been known at least since ancient Egypt. The meat was dried in the sun and wind next to a smoky fire to protect it from insects.

Jerky is sliced meat or strips of meat with the fat trimmed off, spiced by marinating into either sweet or salty liquids. Then dried at low temperature (usually 160⁰F or 70⁰C) or occasionally salted and sun-dried. The result is a ready-to-eat semi-sweet and salty snack which does not need refrigeration to be stored for long periods of time. This product is a nutrient-dense dried meat due to dehydration (1, 25). Beef is the most common meat used to prepare jerky, but recently, meats from other animals also have been used, such as venison, elk, turkey, ostrich, salmon, alligator and tuna. To avoid bacterial growth, the meat must be dried quickly. In order to achieve this, the meat is thinly sliced, or pressed thinly as when ground meat is used.

While jerky can be made by drying the meat in the sun (as natural drying) commercial jerky is made using a food dehydrator which includes: source of heat, air flow to circulate the dry air, trays to hold the food during drying process (33).

Two desirable attributes of jerky are texture and high chewiness which depend on the drying time (23). During the drying process, the use of low heat is needed to avoid cooking or overdrying the meat. A pound of meat or poultry will yield about four ounces of dried jerky (27). The meat is preserved by removing moisture which prevents enzymatic action and microbial growth.

Because of the low moisture content it is unlikely that food borne pathogens would grow in finished beef jerky. In spite of this, some pathogenic bacteria such as *Escherichia coli* O157:H7, *Salmonella*, and *Staphylococcus* have been implicated in some cases of food borne illness outbreaks related to consumption of jerky (22). The objective of this study was to evaluate the microbial safety of beef jerky commercially available in the Stillwater area.

CHAPTER II

Review of Literature

Microbiological safety of jerky

Meat is an excellent culture medium for a wide variety of microorganisms due to its richness in nutrients, good buffering capacity, and moisture content. The micro-flora contained in the meat in addition to the parameters just mentioned make meat extremely perishable (14). Thus, studies to validate food safety on meat products from processing plants are important.

The USDA in May 2004 published a guideline which states the most important issues regarding production of jerky; it was updated on December 2004 (33) and is summarized as follows.

Compliance Guideline for Meat and Poultry Jerky Produced by Small and Very Small Plants

In 2003 there were two points identified by FSIS in making jerky that must be improved by processors. The first was the heat treatment which must be adequate to achieve desired lethality of pathogens. When dry heat is used to dry the meat, it is dried prematurely causing the lethality process to stop. The second point is moisture-protein ratio (MPR) is less important than water activity to determine whether the

process performed was adequate to produce shelf-life stable jerky. Water activity measures the available water contained in a food that can be used for microbial growth. The water activity value provides better confidence of product stability than MPR.

Drying jerky is a method to stabilize the product, but lethality of the heat treatment is needed to produce a safe jerky. The steps to produce a safe jerky are generally as follows:

1. Strip preparation: Ground meat is pressed to form strips or the intact muscle may be sliced into strips.

2. Marination: The meat strips are marinated in an aqueous mixture containing salt, sugar, spices or other flavoring additives.

3. Interventions before drying process: Some antimicrobial interventions are needed to help insure destruction of any pathogenic bacteria present on the meat. Examples of these interventions that may be applied before and after the marination process are:

- Preheating the meat to 71⁰C internally to kill *Salmonella* (17). However, this could adversely affect flavor of the marinade and thus produce an acceptable flavor in the finished product.
- Using a solution of 5% acetic acid, for dipping meat for 10 minutes before marination can reduce the numbers of pathogens, but is not enough to eliminate pathogens (5, 6). This intervention may also produce an undesirable flavor.

4. Lethality of heat treatment: Pathogens such as *Salmonella*, *E. coli* O157:H7, *Staphylococcus aureus*, and *Listeria monocytogenes* must be eliminated. For that purpose, adequate treatment must be applied to eliminate these hazards. The combination of proper time-temperature will help to ensure the safety of the meat

jerky. Relative humidity must be controlled to prevent drying too rapidly. If this is not done properly the degree of lethality is reduced and the bacteria may develop resistance to the heat (9, 12, 15, 16). The relative humidity must be maintained above 90% during the heat process. This may be achieved by steam injection or by using a sealed oven. To use a lower relative humidity the plant must provide documentation proving the heat used at the lower relative humidity is adequate to kill all pathogens in the meat.

5. Drying: After the heat killing process, the jerky should be dried to achieve a water activity of 0.80 or lower.

6. Post-drying heat step: This process may be needed when the heat treatment does not achieve adequate reduction of *Salmonella*. For this, the dried jerky is heated in a 135⁰C (275⁰F) oven for 10 minutes. The *Salmonella* levels will be reduced by approximately 2 logs cycles by this treatment (18).

7. Handling: The sanitation standard operation procedures for the plant should ensure prevention of any cross-contamination or re-contamination of the jerky.

The American Association of Meat Processors (AAMP) in 2004 published a special report regarding the compliance guidelines summarized above, in order to help meat processors understand and comply with USDA requirements (36). The special report explains why some of these requirements may not be satisfactory. One of these requirements is heating the dried jerky in a 135⁰C oven for 10 min. The AAMP said that such temperature may change the desired characteristics of the jerky, and that dryness may not be acceptable in some places of the United States. In addition, most jerky processors may not have a thermal processing oven capable of reaching a dry bulb temperature of 135⁰C. Another requirement is preheating the jerky to 71⁰C in

either water or other marinade solution before drying. The explanation was that most jerky is manufactured, because of leanness, from the bottom round and top round cuts of beef. These cuts have a high concentration of connective tissue and when heated in a solution have the tendency to curl, resulting in a non flat jerky. Furthermore, the internal temperature in jerky may not be checked accurately due to its thinness (4).

Antimicrobial treatments used to minimize pathogens during production of beef jerky

The safety of jerky has been evaluated in several different ways, in order to determine the most effective method to eliminate pathogenic bacteria. The following examples show not only the effectiveness of chemical treatments against target microorganisms, but also physical parameters that favor achievement of such an objective.

Holley (20), using a home-style dehydrator, studied the fate of *Staphylococcus aureus* in marinated and corned beef during the jerky processing. Round steak was sliced and then marinated in a “Great jerky” marinade for 12 h at 4⁰C; corned beef was only sliced without marinating and held for 12 h at 4⁰C. After that, both samples were dipped into the inoculum of *S. aureus*, drained in a beaker, and placed in the dehydrator. The marination ingredients were garlic pepper, garlic salt, brown sugar, soy and Worcestershire sauce, and salt (no amounts specified). The dehydration process was for 4 h at 68⁰C and an additional 4 h at 60⁰C. Eighty five percent of staphylococci were eliminated after 8 h of thermal process and an additional 10% after 7 days of storage at 2.5⁰C.

The rate of inactivation of *E. coli* O157:H7 at two temperatures in dried beef powder adjusted to three different levels of sodium chloride and two different levels of water activities was evaluated by Ryu and others (30). They used commercially available dried beef powder; the inocula were acid-adapted or acid-shocked *E. coli* O157:H7. The *E. coli* was cultured in tryptic soy broth for 16 h then lactic acid was added to the culture to reduce the pH to 4.9 followed by incubation for 2 more h. The sodium chloride contents were 0.5, 3.0, and 20%. The temperatures were 5 and 25⁰C for eight weeks storage. The authors found a higher rate of inactivation at 25⁰C with A_w of 0.34 than at 5⁰C with A_w 0.68. In addition, the dried beef powder containing 20% of sodium chloride exhibited higher level of inactivation of *E. coli* O157:H7 than did those with 0.5 and 3.0%. They suggest that not only the ionic effect of sodium chloride but also the water-binding characteristics, increase death of the bacterial cells. They did not detect protection against osmotic stress or dehydration related by the acid-adaptation or acid-shock mechanisms.

Four methods to process jerky were evaluated by Harrison and others (18) to eliminate *Salmonella*, *E. coli* O157H:7, and *Listeria monocytogenes*. All the samples were beef strips inoculated with the target pathogenic organism (each strain separately). The marinade was a mixture of 60 ml soy sauce, 15 ml Worcestershire sauce, 0.6 g pepper, 1.25 g garlic powder, 1.5 g onion powder, and 4.35 g hickory smoke-flavor salt per 900 g meat. All marination processes were overnight at 4⁰C; and the dehydration process was at 60⁰C. The first method was the traditional one, which consisted of marination, and dehydration. The second method was marination, dehydration, and oven-heating the strips at 135⁰C for 10 minutes. The third method

was marination, boiling the strips in extra marinade for 5 minutes, prior to dehydrating the strips. The fourth method was marination, oven-heating at 163⁰C for 10 minutes, and then dehydrating the strips. The results showed a reduction of 4.6, 5.8, and 3.9-logs for *Salmonella*, *E. coli* O157H:7, and *L. monocytogenes* respectively in treatments 1, 2 and 3. Treatment 4 which included the oven-heating treatment (163⁰C for 10 minutes) after dehydration reduced the numbers of the three pathogenic microorganisms by 2 additional log cycles.

Survival of *E. coli* O157:H7 inoculated into the product was evaluated by Albright and others (2) where four different pre-drying treatments were compared. The drying process was performed into a home-style dehydrator with 19 - 24% of relative humidity for 10 h at 104⁰C followed by storage for 90 days at 21⁰C (room temp.). The four pre-drying treatments were as follows: 1) Immersing the meat into 94⁰C water for 15 s followed by marination; 2) Seasoning in pickling spices for 24 h at 4⁰C, then immersing in pickling brine for 90 s at 78⁰C; 3) Immersing the meat in a solution containing 750 mL of vinegar and 750 mL of water for 20 s at 57.5⁰C followed by marination; 4) Same as number three but in reverse order. All the marination processes were for 24 h at 4⁰C in a solution containing soy sauce, Worcestershire sauce, black pepper, garlic powder, hickory smoke-flavored salt, and onion powder. The pickling spices were a mixture of iodized salt, granulated sugar, and Shilling black pepper. The pickling brine was a solution of water with iodized salt, granulated sugar, and black pepper.

The pickling brine treatment at 78⁰C caused a reduction in the log cfu/cm² of 5.8. The jerky treated in this manner reached an acceptable A_w value by 8 hours of drying. The acceptable range of water activity for beef jerky is < 0.68 in order to have a stable

shelf-life (24). The boiling water and marinating treatment had not reached an acceptable level of water activity value after 10 hours of drying, but it was reached by day 90 of storage. The storage after 10 hours was aerobic at room temperature (21⁰C). This process thus would make the product vulnerable to growth of microorganisms since they had not reached the proper water activity value. The range of pH in beef jerky was from 5.7 to 6.0 which is typical of raw whole beef muscle (26). The authors suggested that the two treatments involving warm (57.5⁰C) vinegar decreased microbial counts due to the acidity of the products.

Raisin extracts and raisin puree were tested as potential antimicrobial additives against *Staphylococcus aureus*, *Escherichia coli* O157:H7, *Listeria monocytogenes* Scott A, *Salmonella choleraesuis*, and *Clostridium perfringens* by Bower and others (3). Raisins were pureed with water (10% w/v) and extracted with ethanol.

Following evaporation of the ethanol the extract was placed on the surface of commercial beef jerky. Jerky strips also were prepared from lean ground beef plus raisin puree at different concentrations. The results showed that the application of extracts increased water activity causing it to become sticky, and also increased microbial growth. Thus the raisin extracts did not have high enough antimicrobial activity at the concentration tested to be of benefit. The authors suggest that adding raisins directly, instead of raisin puree or raisin extracts to the meat, could be done to produce a raisin-jerky product, with improved appearance and mouth feel. However, it would not improve the microbial quality of the product.

The fate of *S. aureus* in several ready-to-eat meat products including beef jerky was evaluated by Ingham and others (21). Four samples of beef jerky were purchased from a local grocery store. The inoculum was prepared as a cocktail using three strains that do not produce enterotoxins. Jerky slices were inoculated with 0.025 mL of the cocktail, allowed to dry for 30 minutes, and then vacuum packaged for storage at 21⁰C for 28 days. The results showed a reduction of 1.0 to 2.6 log cycles by day 7 and 3.2 to 4.5 log cycles by day 28. The authors suggest that the shelf life of beef jerky samples could be considered stable, and that the USDA shelf-stability standards are adequate to prevent pathogenic bacteria growth.

Humidity is an important factor in the drying process as was shown by Harrison and others (19) in comparison of a dehydrator to a smokehouse for drying jerky. In their experiment the relative humidity was controlled at 33% in the smokehouse; while in the dehydrator it was permitted to vary depending on the room air. They concluded that although the dehydrator was effective, the smokehouse was more effective because humidity was better controlled in it. In addition, they evaluated two chemical pretreatments in the marination process. The marination solution contained water, salt, sugar, garlic powder, nitrite, sodium erythorbate, MSG, 4% vinegar, and thyme. Chlorine dioxide (500 and 1200 ppm) and acidic calcium sulfate (1:2 and 1:3 water/calcium sulfate ratios) were the chemicals used. The higher concentration of calcium sulfate was the best method to minimize numbers of *Salmonella spp*, *E. coli* O157:H7, and *L. monocytogenes*.

Outbreaks of food borne illnesses related to jerky

From 1966 to 1995 in New Mexico, 8 outbreaks of gastroenteritis were related to jerky. *Salmonella* and *S. aureus* (six and two cases respectively) were the microorganisms involved (11). Thus, the New Mexico Environment Department planned to evaluate the production processes such as temperature of meat during drying. They developed regulations for the production of jerky such as internal temperature of the meat; those temperatures were 63⁰C for beef, lamb and fish, and 74⁰C for poultry for 3 h (27).

The first case of illness caused by beef jerky recognized in New Mexico since the regulations were implemented was published by Centers for Disease Control and Prevention (8) as follows: The New Mexico Department of Health (NMDOH) detected salmonellosis in two persons during February 1995. Following that an additional 91 cases were identified. The jerky was analyzed and *Salmonella* serotype Montevideo was found. The NMDOH published information advising people not to eat beef jerky from the implicated brand. Five out of the 93 were hospitalized. Cultures were taken from 40 persons 31 of them had *Salmonella sp.*, 12 had *Salmonella montevideo*, and 11 had *Salmonella kentucky*. Jerky samples were obtained from both the manufacturer and from five ill people. Eleven of the 12 beef jerky samples evidently had not been dried properly. In addition, investigators found that neither chemical preservatives nor salt curing were used by jerky producers in New Mexico (11). In August 1995, *Staphylococcus aureus* was found in antelope jerky made at home for private consumption (11).

In 1995 an outbreak of illness caused by *E. coli* O157:H7 in homemade venison jerky was reported in Oregon (22). After investigations, the report concluded that the traditional home-drying treatments were not effective enough to eliminate *E. coli*

O157:H7. It was recommended that precooking venison at 74⁰C before drying would eliminate the organism (27).

Water activity

The minimal moisture content of dried meat required to favor the growth of staphylococci and streptococci is 20%, clostridia 30%, and salmonella 60% (31).

Water activity is an important factor in considering the stability of food, and is also an important parameter that indicates the potential microbial and chemical risk of food (14). Water activity (A_w) is an indicator of the availability of water for chemical and biological reactions. The A_w is calculated by the vapor pressure of water in the head space above a sample in equilibrium in a closed container, divided by the vapor pressure above pure water under the same temperature (7). Currently, we can measure water activity by using a water activity-meter. The US Food and Drug Administration (FDA) as well as similar agencies in other countries, are using A_w as a safety indicator in categorizing food systems (7). In order to reduce A_w of beef products, sodium chloride or other solutes are added to meat during processing (30). Dehydration of beef does not decrease its nutritional value of major components even when stored at 37.7⁰C (1). Vitamin content is other factor that might change during the drying process of beef. There can be a variety of changes depending on the treatment used to dry meat. A study performed by Orent-Keiles and others (28) where retention of thiamine, riboflavin, nicotinic acid, iron, and phosphorous were analyzed, showed that thiamine was the most affected. The degrees of losses in

riboflavin, phosphorous, and even thiamine were in acceptable ranges. Iron content showed an increase which was attributed to contamination from the equipment used in processing meat. The nicotinic acid content did not change.

The content of thiamine at 4.4⁰C was stable, but in contrast, it was not at 21⁰C, and continued to decrease with time. Thiamine loss was detectable, and the loss was almost complete after 10 weeks (37). In addition, they detected that neither niacin nor riboflavin showed any loss, even at 48⁰C and after 42 weeks. The protein quality of beef is slightly reduced by a hot-air drying process, but does not change in freeze-drying (10).

Christen and Jack (7) stated that there are several parameters (e.g. temperature, additives, pH, O₂, and A_w) which must be controlled to avoid not only microbial activity but also chemical reactions causing food deterioration. One of the most important factors is water activity, for which they listed the water activity of some food and its influence on spoilage by microorganisms (Table 1).

Table 2-1. Influence of water activity on microbial growth in various foods^a

| Range of A_w | Microorganisms generally inhibited by lowest a_w in this range | Examples of foods generally within this range of a_w |
|----------------|---|---|
| 0.95 – 1.00 | <i>Pseudomonas</i> , <i>Escherichia</i> , <i>Proteus</i> , <i>Shigella</i> , <i>Klebsiella</i> , <i>Bacillus</i> , <i>Clostridium perfringens</i> , some yeasts | Fresh and canned fruits, vegetables, meat, fish and milk, sausages, breads, foods with up to 40% sucrose or 7% NaCl |
| 0.91 – 0.95 | <i>Salmonella</i> , <i>Vibrio parahaemolyticus</i> , <i>C. botulinum</i> , some molds, <i>Rhodotorula Pichia</i> | Cheeses: Cheddar, Swiss, Muenster, Provolone. Cured ham, fruit concentrates, food with 55% sucrose and 12% NaCl |
| 0.87 – 0.91 | Many yeasts (<i>Candida</i> , <i>Torulopsis</i> , <i>Hansenula</i>), <i>Micrococcus</i> | Salami, sponge cake, dry cheeses, margarine, foods with 65% sucrose and 15% NaCl |
| 0.80 – 0.87 | Most molds (<i>Mycotoxigenic penicillia</i> , <i>Staphylococcus aureus</i> , most <i>Saccharomyces</i> , (<i>bacilli</i>) spp., <i>Debaryomyces</i> | Fruit juice concentrates; sweetened condensed milk; chocolate, fruits, and maple syrups; flour; rice; fruit cake |
| 0.75 – 0.80 | Most halophilic bacteria, mycotoxigenic aspergilli | Jam, marmalade, marzipan, glace fruits, some marshmallows |
| 0.65 – 0.75 | Xerophilic molds (<i>Aspergillus chevalieri</i> , <i>A. candidus</i> , <i>Wallemia sebi</i>), <i>Saccharomyces bisporus</i> | Rolled oats with 10% moisture, grained nougats, fudge, marshmallows, jelly, molasses, raw cane sugar, nuts |
| 0.60 – 0.65 | Osmophilic yeasts (<i>Saccharomyces rouxii</i>), few molds (<i>Aspergillus echinulatus</i> , <i>Monascus bisporus</i>) | Dried fruit containing 15-20% moisture, some toffees and caramels, honey |

^a(Reproduced from Christen and Jack (7))

In conclusion, a food is highly perishable when its A_w falls in a value close to that of pure water (1.0). Those foods which are stable at room temperature are normally in A_w range of 0 - 0.60, with the exception of those commercially sterilized foods (7).

Acidity (pH)

Acidification of meat helps to achieve the removal of moisture during the drying process. Thus, it is more difficult to remove water from meat when it is not acidified due to its higher pH. When the meat is dehydrated it results in the contraction of proteins to expel water (34). The concentration of salts increases during drying process because moisture is evaporated, resulting in an increase in pH and ionic strength (13). A high pH makes the humidity and airflow critical during drying process of a meat product. The control of temperature must be tight because the higher pH makes the product more difficult to dry (34).

An acidic pH will extend shelf life because it eliminates the growth of several microorganisms, and retards the growth of others; “the ultimate pH of meat approaches the isoelectric point of myosin and actomyosin (pH 5.3 - 5.5), the pH at which their net charges is zero (7).” The net charge of a protein will be zero when its isoelectric point (PI) is equal to its pH (14). The result of electrostatic repulsion and net charge reduction is the loss of water-holding capacity and shrinking of myofibrils (7).

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CHAPTER III

MICROBIOLOGICAL EVALUATION OF
COMMERCIAL BEEF JERKY

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ABSTRACT

Beef jerky is a very nutritious and popular ready-to-eat snack which does not need refrigeration for storage. It can, however be contaminated by pathogens before, during, or after processing if the handling methods are not adequate; furthermore, due to minimal processing, the product could still contain some of the micro flora found in raw meat. Although some pathogenic bacteria such as *Escherichia coli* O157:H7, species of *Salmonella*, and *Staphylococcus aureus* have been causes of some cases of food borne illness outbreaks attributed to jerky, not much work has been done to evaluate the microbial safety of beef jerky. The objective of this study was to evaluate the microbial safety of beef jerky commercially available in the Stillwater area.

Forty-two samples of beef jerky were purchased and analyzed. They were aseptically ground and appropriate serial dilutions plated by a spread plate method in duplicate on Baird Parker agar for coagulase positive *Staphylococcus aureus*, and on acidified Potato Dextrose agar for yeast and molds; the pour plate method was used for total plate count on Plate Count Agar, and for coliforms on Violet Red Bile agar. Analyses for *Salmonella* and *E. coli* O157:H7 were done using Minividas equipment following enrichment in Buffered Peptone Water (BPW) for the former and Tryptic Soy Broth and Mac Conkey broth (CT-MAC) for the latter.

No pathogenic bacteria were detected in any sample; the numbers on Plate Count Agar were low. While for a few samples an occasional mold colony appeared on the lowest dilution plated (1:100) most had none. Since we did not detect any pathogens, it is tempting to assume that this product is safe to consume.

INTRODUCTION

Dried meat may contain different levels of microorganisms, which depend on the type of product, its ingredients, and the processing methods (13). There are different reasons to analyze processed food for pathogens. One of these reasons is to determine whether or not it has been contaminated post-processing, which is typically due to exposure of the food to an inadequately sanitized food-processing surface or human contact by food handlers (14). During processing of jerky the reduction of water activity (A_w) is an important step in order to control foodborne pathogens. This is not only done by dehydration but also by adding salts (15). The principal benefit of removing moisture is that enzymes are not able to react with the food nor are bacteria able to grow. Both these effects aid in avoiding spoilage (22).

Some pathogenic bacteria such as *Escherichia coli* O157:H7, species of *Salmonella*, and *Staphylococcus aureus* have been causes of some cases of food borne illness outbreaks attributed to jerky (home made) (6). Some home made jerky producers add neither salt curing nor chemical preservatives; however, they only soak beef strips in marination before the dehydration process (6). There are several different antimicrobial treatments used to minimize pathogens during production of beef jerky. Those treatments have been evaluated to determine the most effective method to achieve such an objective. Commercial production of jerky must follow procedures based on a Hazards Analysis and Critical Control Point program to ensure destruction of *E. coli* O157:H7, *Salmonella*, and *Listeria monocytogenes* (23). The Food Safety Inspection Services of USDA (FSIS-USDA) has identified two points where the jerky processors need to improve methodology; the first is the heat treatment which must be

adequate to achieve desired lethality of pathogens; the second was relying on moisture –protein-ratio (MRP) rather than water activity (23).

Ryu and others (19), evaluated the rate of inactivation of *E. coli* O157:H7 at two temperatures in dried beef powder adjusted to three different levels of sodium chloride and two different levels of water activity. They suggested that the death of bacterial cells was favored for both low moisture level and the ionic effect of sodium chloride.

Harrison and other (9), evaluated four methods to process jerky in order to eliminate pathogenic bacteria. They combined marinating, drying, and heating treatments for destruction of *Salmonella*, *Escherichia coli* O157:H7, and *Listeria monocytogenes*, resulting in a reduction of 4.6, 5.8, and 3.9 logs respectively. Since humidity is an important factor in the drying process, Harrison and others (10), did compare the use of dehydrator versus a smokehouse for drying jerky. In addition, they treated the samples with two different chemicals; chlorine dioxide and calcium sulfate at two different concentrations. The best method to minimize *Salmonella*, *E. coli* O157:H7, and *L. monocytogenes* was that containing calcium sulfate (1:3 water:calcium sulfate ratio).

The objective of this study was to evaluate the microbial quality of commercially processed beef jerky available in Stillwater, Oklahoma.

MATERIALS AND METHODS

Preparation of samples

Forty-two samples of beef jerky were purchased from retail stores in the Stillwater area, including 11 different brands. For multiple samples from a single brand each had different a lot number. They were kept at room temperature (25⁰C) in the darkness until analyzed. The samples were aseptically ground in a household Universal food grinder. The ground sample was mixed and divided into five parts: 25 g to perform tests for *Salmonella* tests; 25 g to perform tests for *E. coli* O157:H7 tests; 11 g to make serial dilutions for plate counts for total counts, coagulase positive staphylococci, yeast and molds, and coliforms; other portions were used for measurement of water activity and pH. The four portions for microbial analyses were placed separately into sterile stomacher filter bags (Nasco whirl-pack, Fort Atkinson, WI). The initial dilution for plate counts was prepared by adding 99 ml of sterile diluent (0.1% sterile peptone water) to the 11 g portion in the stomacher bag and pummeling it for 2 minutes in a stomacher (Stomacher 400, Seward Medical Ltd., London, United Kingdom) to obtain a 1:10 dilution.

Procedures for Microbial Analyses

Total aerobic plate count

Appropriate serial dilutions were prepared from the initial 1:10 dilution of the sample and plated by the pour plate method with Plate Count Agar (PCA, Difco™, Sparks, Md., U.S.A.). One milliliter of sterile 0.5% (w/v) TTC (2, 3, 5-triphenyl tetrazolium chloride, Sigma, St. Louis, Mo., U.S.A.) was added to 100 mL of PCA just prior to pouring plates in order to make the colonies visible and distinguishable from the particles of the samples. Once the plates solidified, they were incubated for 48 h at 32⁰C. After incubation, colonies were counted with the aid of a Leica Quebec Darkfield Colony Counter (Leica, model 3324, Buffalo, NY). Results were expressed as colony forming units per gram (cfu/g).

Yeast and Molds

Yeasts and Molds were enumerated on Potato Dextrose Agar (PDA, Difco™). Just prior to using the medium, it was acidified (to pH 3.5) by adding 1.8 mL of sterile 10% tartaric acid, (Fisher Scientific, Pittsburgh, PA., USA) to each 100 mL of PDA that had been melted and tempered to 45⁰C. The PDA plates were poured, solidified, and dried overnight before being inoculated. The desired dilutions were spread plated in duplicate onto the PDA plates. All the plates were incubated in an upright position at room temperature (23⁰C ± 2) for 5 days.

Coliforms

Violet Red Bile Agar (VRBA, Difco™) was used to analyze jerky samples for coliforms. Appropriate dilutions were plated in duplicate with VRBA by a pour plate method. Once solidified, the plates were overlaid with VRBA and incubated at 37°C for 48 hours. Typical coliform colonies were counted (purple-red, ≥ 1 mm diameter and surrounded by a zone of precipitated bile acids).

Coagulase positive staphylococci

The samples were plated onto the Baird-Parker agar (Difco™) containing 10% egg yolk (EY Tellurite Enrichment, Difco™) by the spread-plate method. One milliliter from each 10 fold dilution was plated in measured volumes (0.3, 0.3, and 0.4 mL respectively) on three plates of solidified agar. The plates were kept in an upright position until the liquid was absorbed by the agar, then were inverted and incubated at 37°C for 48 hours. Typical colonies (jet black to dark gray, smooth, convex, with entire margins, and surrounded by an opaque zone or a clear halo) were counted for determining counts. If typical colonies were detected they were tested for coagulase (24).

Test for *Escherichia coli* O157:H7

Tryptic soy broth (225 mL) (TSB, Difco™) supplemented with 10% novobiocin (novobiocin antimicrobial supplement, Difco™) was added to the stomacher bags that contained 25g of sample. The bags were pummeled for 2 minutes and incubated at

41⁰C for 6-7 hours. After incubation, 1 mL of the enrichment culture was transferred into 9 mL of MacConkey broth (DifcoTM) supplemented with cefixime (0.05 mg/L) and potassium tellurite (2.5 mg/L) (CT-MAC, Dynal[®], Lake Success, NY) and incubated at 37⁰C for 18 hours. One mL of the resulting culture was transferred into a sterile tube, sealed and heated at 100⁰C for 15 minutes; meanwhile CT_MAC cultures were kept in the refrigerator for later use if needed. After being cooled, the heated samples were analyzed serologically for *E. coli* O157 by miniVIDAS assay (miniVIDAS procedure is explained in Appendix A; miniVIDAS, BioMérieux, Florence, Italy)

Unheated portions CT_MAC cultures of those samples that yielded positive results in the miniVidas assay were immuno-concentrated by using ICE- kits (for mini-VIDAS). Upon completion of the ICE, using a sterile micropipette the immuno-concentrated sample from the kit was plated by a spread plate method on the surface of Sorbitol Mac Conkey agar supplemented with MUG (4-methylumbilliferyl-beta-D-gluconic acid) (SMAC, DifcoTM; MUG, Sigma) and on chromagenic (CHROMagarTM, Paris, France) agar for isolation of typical colonies. The plates were incubated at 37⁰C for 24 hours. Plates were examined for typical colonies of *E. coli* O157 under an ultraviolet lamp (VWRTM Scientific, Tonawanda, NY) in a darkened area. Any typical colonies were tested for agglutination reaction by using Remel Solutions (Remel, RIMTM, Lenexa, KS). Identity of isolates from positive samples was confirmed using the Vitek system (VITEK 32, BioMérieux, Hazelwood, MO) (VITEK procedure is explained in Appendix A). Positive control samples were included.

Test for *Salmonella*

The enrichment culture for *Salmonella* detection was made by adding 225 mL of Buffered Peptone Water (BPW, see the recipe in Appendix A) to the remaining stomacher bag containing 25 g of sample and pummeling for 2 minutes. Then the samples were incubated at 37⁰C for 24 h. An aliquot (800 µL) of the BPW enrichment culture was transferred to VIDAS strips for immuno-concentration of *Salmonella* (ICS) following the manufacturer's instructions. Upon completion of the ICS, 400 µL were transferred into a 2 mL of ICS broth previously prewarmed at 41⁰C for 30 minutes and incubated at 41⁰C for 6 hours. After the incubation, 1 mL from the ICS was transferred into a sterile tube, sealed, and heated at 100⁰C for 15 minutes. The remainder of the ICS tube was kept in the refrigerator to use later if needed. The heated ICS broth culture (500 µL) was transferred to the VIDAS for serological detection of *Salmonella* following the instruction of the VIDAS SLM package and mini-VIDAS system. The unheated portion of the ICS cultures which yielded positive results were streaked onto XLD (xilose-lysine-deoxycholate, DifcoTM) and HEKTOEN (DifcoTM) agars for confirmation. The plates were inverted and incubated at 37⁰C for 24 hours to isolate typical colonies which were identified by the VITEK system.

Acidity (pH) measurement

A 1:10 dilution (w/v) was prepared from each ground jerky sample with deionized water and pummeled in a stomacher for 2 minutes. A model AB15 Accumet[®] pH-meter (Fisher Scientific, Pittsburg, PA USA) was used to measure acidity (pH).

Measurement of water activity (Aw)

Samples were ground in a food processor (Cuisinart Mini-Prep Plus Model DLC-2, UL, USA), within 1-2 minutes a small amount of it was used to measure water activity by using a model CX2 Aqualab (Decagon Devices, Inc., Pullman, Wash., USA). The measurements were made on duplicate samples.

Statistical Analysis

The total counts (cfu/g) of aerobic bacteria were converted to log₁₀ cfu/g and analyzed by Statistical Analysis System (SAS 2006, software version 9.1, Cary, North Carolina, USA). The samples were grouped by flavor and their data analyzed for least significant difference (LSD) by t Test. The same t Test analysis was applied for comparison between brands. Least square means were separated by Fisher's least significance difference (LSD) among brands and flavors using the general linear models (GLM) procedure of SAS. Significant differences were assumed at $P < 0.05$ for all statistical analyses. The statistical model (6) used was the following:

$$Y_i = a + b \text{ pH} + c \text{ Aw} + d \text{ Brand} + e \text{ Flavor} + \text{error}$$

$$Y_{ijklm} = a + b_i \text{ pH} + c_j \text{ Aw} + d_k \text{ Brand} + l_l \text{ Flavor} + \epsilon_{ijklm}$$

i = number of pH

j = number of water activity

k = number of brand

l = number of flavor

m = replication

RESULTS AND DISCUSSION

No pathogenic bacteria such as *E. coli* O157:H7, *Salmonella*, and *S. aureus* were detected in any of the 42 samples tested. While for a few samples an occasional mold colony appeared on the lowest dilution plated (1:100) most had none. Leistner (16) and Horner & Anagnostopoulos (12) concluded that, the growth of molds in dried meat products is inhibited by lower pH and low A_w . When yeasts and molds are not visibly present in the plates, it does not mean that they are not present (18). Instead, it may mean that the numbers were low and were not detectable in the smallest dilution which was 1:100. (The whole raw data is in appendix B, where the results showed no presence of coliforms.) The negative results for pathogens, coliforms and yeasts and molds indicate high microbial quality and safety. Their absence suggests that the antimicrobial treatments used in processing jerky were effective. Antimicrobial treatments used to prepare jerky have been reported to have significant effect on microbial survival (3). The combination of preservatives, pH, oxidation-reduction potential, temperature, and A_w to inhibit microorganisms in foods is known as the hurdle effect (17).

The results for counts of aerobic bacteria, water activity, and pH are reported in the following 5 tables. The jerky samples in each table represent different flavors of jerky.

Table 3-1. Total plate counts of from original flavor jerky.

| Sample | Brand ^{aa} | Log ₁₀ cfu/g ^a | Water activity (Aw) ^a | Acidity (pH) ^a |
|-----------|---------------------|--------------------------------------|----------------------------------|---------------------------|
| JERKY# 3 | B | 3.11 | 0.69 | 5.1 |
| JERKY # 4 | C | 2.77 | 0.65 | 5.4 |
| JERKY # 5 | D | 1.00 | 0.69 | 5.8 |
| JERKY # 6 | A | 1.00 | 0.69 | 5.6 |
| JERKY #11 | A | 4.69 | 0.74 | 5.8 |
| JERKY #14 | C | 3.71 | 0.75 | 5.7 |
| JERKY #17 | F | 2.41 | 0.60 | 5.6 |
| JERKY #19 | F | 2.28 | 0.63 | 5.6 |
| JERKY #21 | B | 4.18 | 0.74 | 5.3 |
| JERKY #25 | C | 2.20 | 0.75 | 5.6 |
| JERKY #27 | G | 1.60 | 0.75 | 5.8 |
| JERKY #29 | I | 1.30 | 0.68 | 6.0 |
| JERKY #30 | H | 1.00 | 0.81 | 5.6 |
| JERKY #32 | K | 3.60 | 0.70 | 5.3 |
| JERKY #33 | J | 1.00 | 0.69 | 6.0 |
| JERKY #35 | I | 1.70 | 0.64 | 6.0 |
| JERKY #36 | A | 1.00 | 0.78 | 5.9 |
| JERKY #41 | A | 1.00 | 0.76 | 5.9 |
| Mean | | 2.19 | 0.71 | 5.7 |

^aEach value represents results from analysis of one sample.

^{aa}The letters represent different brand of jerky.

Table 3-2. Total plate counts of from teriyaki flavor jerky.

| Sample | Brand ^{aa} | Log ₁₀ cfu/g ^a | Water activity (Aw) ^a | Acidity (pH) ^a |
|-----------|---------------------|--------------------------------------|----------------------------------|---------------------------|
| JERKY # 7 | D | 1.00 | 0.66 | 5.8 |
| JERKY # 9 | C | 1.00 | 0.72 | 5.5 |
| JERKY #10 | A | 1.00 | 0.70 | 5.9 |
| JERKY #15 | A | 4.69 | 0.61 | 5.6 |
| JERKY #26 | H | 1.00 | 0.70 | 5.8 |
| JERKY #31 | J | 1.00 | 0.64 | 6.0 |
| JERKY #39 | A | 1.00 | 0.74 | 5.8 |
| JERKY #40 | A | 1.00 | 0.74 | 5.8 |
| JERKY #24 | G | 2.61 | 0.82 | 5.9 |
| Mean | | 1.59 | 0.70 | 5.8 |

^aEach value represents results from analysis of one sample.

^{aa}The letters represent different brand of jerky.

Table 3-3. Total plate counts of from smoked flavor jerky.

| Sample | Brand ^{aa} | Log ₁₀ cfu/g ^a | Water activity (Aw) ^a | Acidity (pH) ^a |
|-----------|---------------------|--------------------------------------|----------------------------------|---------------------------|
| JERKY # 1 | A | 1.00 | 0.63 | 5.8 |
| JERKY # 2 | A | 2.04 | 0.79 | 5.5 |
| JERKY #37 | A | 2.34 | 0.69 | 5.9 |
| JERKY #38 | A | 1.00 | 0.69 | 5.9 |
| Mean | | 1.59 | 0.70 | 5.8 |

^aEach value represents results from analysis of one sample.

^{aa}The letters represent different brand of jerky.

Table 3-4. Total plate counts of from peppered flavor jerky.

| Sample | Brand ^{aa} | Log ₁₀ cfu/g ^a | Water activity (Aw) ^a | Acidity (pH) ^a |
|-----------|---------------------|--------------------------------------|----------------------------------|---------------------------|
| JERKY # 8 | E | 1.00 | 0.71 | 5.7 |
| JERKY #12 | D | 3.00 | 0.70 | 6.1 |
| JERKY #13 | A | 1.00 | 0.71 | 5.8 |
| JERKY #16 | C | 1.00 | 0.72 | 5.8 |
| JERKY #22 | B | 3.44 | 0.70 | 5.5 |
| JERKY #23 | G | 3.52 | 0.79 | 5.4 |
| JERKY #34 | I | 1.00 | 0.69 | 5.6 |
| JERKY #28 | I | 1.48 | 0.64 | 5.9 |
| Mean | | 1.93 | 0.71 | 5.7 |

^aEach value represents results from analysis of one sample.

^{aa}The letters represent different brand of jerky.

Table 3-5. Total plate counts of from hot & spicy flavor jerky.

| Sample | Brand ^{aa} | Log ₁₀ cfu/g ^a | Water activity (Aw) ^a | Acidity (pH) ^a |
|-----------|---------------------|--------------------------------------|----------------------------------|---------------------------|
| JERKY #18 | F | 2.45 | 0.61 | 5.6 |
| JERKY #20 | F | 2.30 | 0.63 | 5.5 |
| JERKY #42 | A | 2.15 | 0.76 | 6.0 |
| Mean | | 2.3 | 0.67 | 5.7 |

^aEach value represents results from analysis of one sample.

^{aa}The letters represent different brand of jerky.

Table 3-6. Means of total counts of aerobic bacteria by brand.

| Brand ¹ | Number of samples | Mean of Log ₁₀ cfu/g ² |
|--------------------|-------------------|---|
| A | 14 | 1.78 ^a |
| B | 3 | 3.58 ^{bcf} |
| C | 5 | 2.14 |
| D | 3 | 1.67 ^d |
| E | 1 | 1.00 |
| F | 4 | 2.36 |
| G | 3 | 2.58 |
| H | 2 | 1.00 ^d |
| I | 4 | 1.37 ^d |
| J | 2 | 1.00 ^{dg} |
| K | 1 | 3.60 |

¹The letters represent different brand of jerky.

²Each value represents mean of aerobic bacteria of one brand.

Different super script letters show a significant difference at 5% level

(All the counts of aerobic bacteria were statistically analyzed. Analysis results can be seen in the appendix D.)

From the statistical model, it was observed that the only parameter which indicated a significant relationship in the total counts of aerobic bacteria was pH. This means that differences in pH may be related to the numbers of the aerobic bacteria. The aerobic growth of bacteria is lower if the acidity increases (decreasing pH value) since acidity can influence bacteria growth. Acidity also may influence the bactericidal action of the heating or drying process. Microbial growth depends on the nature of proteins and is greatly influenced by pH which also affects the Aw requirements of microorganisms (8).

In addition to the global analysis using the statistical model, data analyses were conducted further to determine whether or not there were any other significant

differences amongst samples' brands and flavors. Data analyses that were conducted are the t Test and least squares means. Even though the model did not give any significant differences for parameters other than pH, when the t Test was performed in the brand comparison analysis, some significant differences were observed for comparison between brand B and some other brands (brand A, D, I, H, and J). This suggests that brand B has better microbial quality than brands A, D, I, H, and J. However, the least squares means revealed no significant differences for brand comparison. No were significant differences observed using t Test. This data shows that flavoring did not differentiate the quality of the products.

All the microorganisms have a minimum, optimum, and maximum acidity and water activity for growth (8); a water activity of 0.85 is considered the safe cutoff for pathogen growth which is based on the minimum water activity needed for *S. aureus* toxin production (7). If the water activity is below 0.70 in jerky, pathogens will not survive vacuum packaged storage at ambient temperature (2). Although some pathogenic bacteria such as *Escherichia coli* O157:H7, species of *Salmonella*, and *Staphylococcus aureus* have been causes of some cases of food borne illness outbreaks attributed to jerky (home made), not much work has been done to evaluate the microbial safety of beef jerky (6). Some home made jerky producers add neither salt curing nor chemical preservatives; however, they only soak beef strips in marination before dehydration process (6).

CONCLUSION

Since we did not detect any pathogens, it is tempting to assume that these commercially available jerky are safe to consume. This could be because the processing preparation for jerky was adequate to eliminate them. However, the samples were not tested for enterotoxins or mycotoxins. In addition, the final water activity and pH values for storage were in the acceptable ranges in order to avoid any bacterial growth. Vacuum packing for storage of beef jerky is another important process that could be adapted to prevent microbial growth. Since jerky is low moisture content people assume that there are no microbiological problems.

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APPENDIX A

MiniVIDAS principle and procedure:

Each mini VIDAS assay kit provides the materials required to run a specific assay.

The materials vary for each assay, but generally a kit contains:

Single or Dual Reagent Strips

SPRs (Solid Phase Receptacle) One per Single Reagent Strip and Two per Dual

Reagent Strip

Controls

Standards, as required

Sample treatment reagents, as required

A package insert

Master Lot Data Card or TOS Sheet

Single Reagent Strip (where the sample is placed): It has ten wells. The first one is an empty well in which to place the sample. The next eight wells contain reagents or washes. The last well is an optical cuvette where the substrate reaction is measured from its fluorescent reading.

Solid Phase Receptacle block is part of an automated pipetting system that uses the SPRs to mix and transfer reagents during processing. The mini VIDAS uses the SPR to withdraw or dispense liquid from or to a well in the Reagent Strip. The beveled tip of the SPR enables it to pierce the protective seal that covers the wells in a Reagent Strip. The Reagent Strip tray then moves in and out to allow liquids to be transferred

from one well to another. The mini VIDAS uses the SPR to accomplish all the required processing steps, including sampling, mixing, and washing. For example:

- The sample is drawn into and out of the SPR.
- The target analyte from the sample binds to the SPR's interiors coating (antibody, antigen, etc.).
- Various washes remove unbound and interfering substances.
- The target analyte is labeled by an enzyme-conjugated antibody, forming a "sandwich."
- The immobilized enzyme catalyzes the breakdown of the substrate into a fluorescent end product.
- The optical scanner obtains fluorescence reading of the substrate material and transmits the measurement to the central processing unit for once reading of the substrate material and transmits the measurement to the central processing unit for analysis.

Procedure:

Type the code test on the instrument. The standard must be run and identified by "S1" for the sample ID. Mix the standard and samples before use in order to improve reproducibility. Pipette precisely 800 μL of standard in the strip identified as S1; the same amount of each sample into well number 4 of the strips. Insert the strips into the instrument and initiate the assay steps as directed in the Operator's Manual. The amount of sample can vary if it is running for assay or immunoconcentration.

VITEK procedure:

1. Fill test tube with prepared diluted sample
2. Place test tube in a single tube filling stand
3. Fit a bent transfer tube into the inlet port
4. Complete test kit. Card is vertical
5. Fill the card by placing it in the filler's vacuum-chamber
6. Remove transfer tube
7. Seal the port
8. Place inoculated card into carousel tray
9. Load the tray into the Reader/Incubator
10. Interim results
11. Read final results in the computer.

BUFFERED PEPTONE WATER (Difco manual)

INTENDED USE

Buffered peptone water is a pre-enrichment medium used for increasing recovery of injured *Salmonella* species from foods prior to selective enrichment and isolation.

PRINCIPLES

Sublethal injury to salmonellas may result fro food preservation techniques involving heat, desiccation, preservatives, and high osmotic pressure or pH changes (5).

Enriching injured cells in lactose broth (pH= 6.9 ± 0.2) may be further detrimental to their recovery (1). Preenrichment with buffered peptone water (pH= 7.2 ± 0.2)

insures maintaining a high pH over the 24-hour incubation period, resulting in repair

of cells that may have an increased sensitivity to low pH (20). This is particularly important for vegetables specimens which have a low buffering capacity.

FORMULA BACTO BUFFERED PEPTONE WATER

Ingredients per liter

| | |
|-------------------------------------|--------------------|
| Peptone..... | 10 g (Difco™) |
| Sodium Chloride..... | 5 g (Sigma) |
| Sodium Phosphate, Dibasic..... | 3.5 g (Sigma) |
| Potassium Phosphate, Monobasic..... | 1.5 g (EM Science) |

Final pH 7.2 ± 0.2 at 25°C

Sterilize in the autoclave for 15 minutes at 15 lbs pressure (121°C)

APPENDIX B

RAW DATA OF MICROBIAL COUNTS IN JERKY SAMPLE

The media where the samples were inoculated are abbreviated as follow

PCA (Plate count agar) used to enumerate total aerobic bacteria.

PDA (Potato dextrose agar) used to enumerate yeasts and molds.

VRBA (Violet red bile agar) used to enumerate coliforms.

BPA (Baird Parker agar) Used to enumerate coagulase positive staphylococci

| Sample | PCA | PDA | VRBA | BPA |
|------------|-----------|-----------|-----------|-----------|
| JERKY # 1 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 2 | 1.10E+02 | 1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 3 | 1.30E+03 | 1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 4 | 5.90E+02 | 1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 5 | <1.00E+01 | 1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 6 | 1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 7 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 8 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 9 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 10 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 11 | 4.90E+04 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 12 | 1.00E+03 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 13 | 1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 14 | 5.10E+03 | <1.00E+02 | <1.00E+01 | <1.00E+01 |

| | | | | |
|------------|-----------|-----------|-----------|-----------|
| JERKY # 15 | 4.90E+04 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 16 | 1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 17 | 2.60E+02 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 18 | 2.80E+02 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 19 | 1.90E+02 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 20 | 2.00E+02 | 1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 21 | 1.50E+04 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 22 | 2.80E+03 | 1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 23 | 3.30E+03 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 24 | 4.10E+02 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 25 | 1.60E+02 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 26 | <1.00E+01 | 1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 27 | 4.00E+01 | 1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 28 | 3.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 29 | 2.00E+01 | 1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 30 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 31 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 32 | 4.00E+03 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 33 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 34 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 35 | 5.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 36 | 1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 37 | 2.20E+02 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 38 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 39 | 1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |

| | | | | |
|------------|-----------|-----------|-----------|-----------|
| JERKY # 40 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 41 | <1.00E+01 | <1.00E+02 | <1.00E+01 | <1.00E+01 |
| JERKY # 42 | 1.40E+02 | <1.00E+02 | <1.00E+01 | <1.00E+01 |

APPENDIX C

DESCRIPTION OF JERKY SAMPLES

Jerky samples divided by brand

| BRAND ID CODE | SAMPLE CODE | FLAVOR (Code) | INGREDIENTS |
|---------------|-------------|--------------------------------|--|
| A | 1 | Original Mesquite-smoked (SMO) | Beef, water, sugar, soy sauce (water, wheat, soybeans, salt), salt, corn syrup, flavorings, hydrolyzed corn gluten, dextrose, paprika, monosodium glutamate, sodium erythorbate, sodium nitrite. |
| | 2 | Original Mesquite-smoked (SMO) | Spices, brown sugar, dried beef stock, maltodextrin, smoke flavor. |
| | 6 | Original (ORI) | Without smoke flavor |
| | 10 | Teriyaki (TER) | Fructose, hydrolyzed corn protein, paprika extract. |
| | 11 | Original | |
| | 13 | Peppered (PEP) | Black pepper. |
| | 15 | Teriyaki | |
| | 36 | Pepperoni (ORI) | Flavorings, spices, paprika extract, citric acid. |
| | 37 | Hickory smoked (SMO) | |
| | 38 | Hickory smoked | |
| | 39 | Teriyaki | |
| | 40 | Teriyaki | |
| | 41 | Original | |

| | | | |
|----------|----|-------------------|--|
| | 42 | Sweet & hot (HOT) | |
| B | 3 | Original | Beef, corn syrup solids, dextrose, hydrolyzed corn and soy protein, salt, natural smoke flavor, flavorings, water, vinegar, sugar, molasses, sodium erythorbate, caramel color, citric acid, sodium nitrite. |
| | 21 | Original | |
| | 22 | Peppered | Vinegar, molasses. |
| C | 4 | Original | Beef, water, sugar, vinegar, salt, maltodextrin, flavorings, monosodium glutamate, spice, sodium erythorbate, citric acid, partially hydrogenated soybean oil, sodium nitrite. |
| | 9 | Teriyaki | Soy sauce powder (soybeans, salt, wheat), hydrolyzed corn protein, paprika, disodium inosinate |
| | 14 | Original | |
| | 16 | Peppered | |
| | 25 | Original | |
| D | 5 | Original | Beef, water, brown sugar, salt, less than 2% of hydrolyzed corn and wheat proteins, flavor, sodium nitrite, maltodextrin, yeast extract, malic acid, soy lecithin. |
| | 7 | Teriyaki | Soy sauce (water, wheat, soybeans, salt), wine, vinegar, garlic, succinic acid, corn syrup solids, lemon juice solids. |
| | 12 | Peppered | Black pepper. |
| | 8 | Peppered | Beef, water, brown sugar, salt, hydrolyzed soy protein, |

| | | | |
|----------|----|--------------------------------------|---|
| E | | | dextrose, flavorings, soy sauce (water, protein extracts from soybeans, salt, corn syrup, caramel color), natural smoke flavor, monosodium glutamate, sodium erythorbate, sodium nitrite. |
| F | 17 | Original | Beef, Worcestershire sauce (distilled vinegar, molasses, corn syrup, water, salt, caramel color, garlic powder, sugar, spices, anchovies, tamarind, natural flavor), salt, soy sauce (water, protein extracts from soybeans, salt, corn syrup, caramel color, potassium sorbate), flavorings, salt, monosodium glutamate, onion, garlic, sodium nitrite, bromelain. |
| | 18 | HOT-N-SPICY (HOT) | |
| | 19 | Original | |
| | 20 | HOT-N-SPICY | |
| G | 23 | Peppered (tenderized with Bromelain) | Beef, water, sugar, salt, corn syrup solids, salt, maltodextrin, dried soy sauce (wheat, soybeans, salt), monosodium glutamate, flavoring, hydrolyzed corn protein, spice, sodium erythorbate, paprika, smoke flavor, sodium nitrite. |
| | 24 | Teriyaki (tenderized with Bromelain) | |
| | 27 | Original (tenderized with Bromelain) | |
| H | 26 | Teriyaki | Beef, water, soy sauce (water, wheat, soybeans, salt) less than 2% of: monosodium glutamate, salt, maltodextrin, wine, flavoring, sugar, vinegar, brown sugar, lemon juice solids, succinic acid, sodium erythorbate, yeast extract, |

| | | | |
|----------|----|---------------------|---|
| | | | tamarind extractives, sodium nitrite, paprika extractives, citric acid, caramel color, soy lecithin. |
| | 30 | Original | |
| I | 28 | Peppered | Beef, brown sugar, water, salt, papaya juice, black pepper, vinegar, garlic powder, monosodium glutamate, citric acid, sodium nitrite. |
| | 29 | Brown sugar (ORI) | |
| | 34 | Peppered | |
| | 35 | Old fashioned (ORI) | |
| J | 31 | Teriyaki | Beef seasoning (soy sauce [water, soybeans, wheat, salt], sugar, sake [water, glucose, sweet rice extract, salt, lactic acid, succinic acid] water, natural flavorings), water, brown sugar, seasoning blend (soy sauce powder [soy sauce {wheat, soybeans, salt}, maltodextrin, salt], flavor [maltodextrin, flavor, sulfur dioxide], brown sugar, spices, onion, garlic), salt, sodium erythorbate, sodium nitrite. |
| | 33 | Original | |
| K | 32 | Original | |

APPENDIX D

STATISTICAL DATA

```

data bacteria;
  input JerkyNum pH Aw T Lcfu Brand$ Flavor$ @@;
  datalines;
1 5.80 0.63 22.90 1.00 A SMO
2 5.50 0.79 23.00 2.04 A SMO
3 5.10 0.69 23.00 3.11 B ORI
4 5.40 0.65 22.80 2.77 C ORI
5 5.80 0.69 22.80 1.00 D ORI
6 5.60 0.69 22.80 1.00 A ORI
7 5.80 0.66 21.80 1.00 D TER
8 5.70 0.71 22.20 1.00 E PEP
9 5.50 0.72 22.00 1.00 C TER
10 5.90 0.70 22.40 1.00 A TER
11 5.80 0.74 22.90 4.69 A ORI
12 6.10 0.70 23.00 3.00 D PEP
13 5.80 0.71 22.90 1.00 A PEP
14 5.70 0.75 23.00 3.71 C ORI
15 5.60 0.61 23.50 4.69 A TER
16 5.80 0.72 23.30 1.00 C PEP
17 5.60 0.60 23.10 2.41 F ORI
18 5.60 0.61 22.70 2.45 F HOT
19 5.60 0.63 22.90 2.28 F ORI
20 5.50 0.63 23.30 2.30 F HOT
21 5.30 0.74 23.30 4.18 B ORI
22 5.50 0.70 23.50 3.44 B PEP
23 5.40 0.79 23.00 3.52 G PEP
24 5.90 0.82 23.20 2.61 G TER
25 5.60 0.75 23.30 2.20 C ORI
26 5.80 0.70 22.70 1.00 H TER
27 5.80 0.75 23.10 1.60 G ORI
28 5.90 0.64 23.30 1.48 I PEP
29 6.00 0.68 23.50 1.30 I ORI
30 5.60 0.81 23.10 1.00 H ORI
31 6.00 0.64 23.20 1.00 J TER
32 5.30 0.70 23.20 3.60 K ORI
33 6.00 0.69 23.10 1.00 J ORI
34 5.60 0.69 23.30 1.00 I PEP
35 6.00 0.64 23.30 1.70 I ORI
36 5.90 0.78 23.00 1.00 A ORI
37 5.90 0.69 23.00 2.34 A SMO
38 5.90 0.69 23.00 1.00 A SMO
39 5.80 0.74 23.00 1.00 A TER
40 5.80 0.74 23.20 1.00 A TER
41 5.90 0.76 23.20 1.00 A ORI
42 6.00 0.76 23.00 2.15 A HOT
;
proc glm data=bacteria;
  class Brand Flavor;
  model Lcfu = pH Aw Brand Flavor;
  lsmeans Brand flavor/tdiff;
run;

```


The GLM Procedure

Class Level Information

| Class | Levels | Values |
|--------|--------|-----------------------|
| Brand | 11 | A B C D E F G H I J K |
| Flavor | 5 | HOT ORI PEP SMO TER |

Number of Observations Read 42
Number of Observations Used 42

The GLM Procedure

Dependent Variable: Lcfu

| Source | DF | Sum of Squares | Mean Square | F Value | Pr > F |
|-----------------|----|----------------|-------------|---------|--------|
| Model | 16 | 21.00170787 | 1.31260674 | 1.03 | 0.4640 |
| Error | 25 | 31.96850403 | 1.27874016 | | |
| Corrected Total | 41 | 52.97021190 | | | |

| | | | |
|----------|-----------|----------|-----------|
| R-Square | Coeff Var | Root MSE | Lcfu Mean |
| 0.396481 | 57.51990 | 1.130814 | 1.965952 |

| Source | DF | Type I SS | Mean Square | F Value | Pr > F |
|--------|----|-------------|-------------|---------|--------|
| pH | 1 | 11.27211549 | 11.27211549 | 8.82 | 0.0065 |
| Aw | 1 | 0.00134174 | 0.00134174 | 0.00 | 0.9744 |
| Brand | 10 | 8.51528041 | 0.85152804 | 0.67 | 0.7445 |
| Flavor | 4 | 1.21297023 | 0.30324256 | 0.24 | 0.9147 |

| Source | DF | Type III SS | Mean Square | F Value | Pr > F |
|--------|----|-------------|-------------|---------|--------|
| pH | 1 | 0.35184819 | 0.35184819 | 0.28 | 0.6045 |
| Aw | 1 | 0.71610695 | 0.71610695 | 0.56 | 0.4612 |
| Brand | 10 | 8.56584724 | 0.85658472 | 0.67 | 0.7411 |
| Flavor | 4 | 1.21297023 | 0.30324256 | 0.24 | 0.9147 |

The GLM Procedure

t Tests (LSD) for Lcfu

NOTE: This test controls the Type I comparisonwise error rate, not the experimentwise error rate.

| | |
|--------------------------|---------|
| Alpha | 0.05 |
| Error Degrees of Freedom | 25 |
| Error Mean Square | 1.27874 |
| Critical Value of t | 2.05954 |

Comparisons significant at the 0.05 level are indicated by ***.

| Brand Comparison | Difference Between Means | 95% Confidence Limits | | |
|------------------|--------------------------|-----------------------|--------|-----|
| K - B | 0.0233 | -2.6659 | 2.7126 | |
| K - G | 1.0233 | -1.6659 | 3.7126 | |
| K - F | 1.2400 | -1.3639 | 3.8439 | |
| K - C | 1.4640 | -1.0872 | 4.0152 | |
| K - A | 1.8207 | -0.5900 | 4.2314 | |
| K - D | 1.9333 | -0.7559 | 4.6226 | |
| K - I | 2.2300 | -0.3739 | 4.8339 | |
| K - H | 2.6000 | -0.2524 | 5.4524 | |
| K - J | 2.6000 | -0.2524 | 5.4524 | |
| K - E | 2.6000 | -0.6936 | 5.8936 | |
| B - K | -0.0233 | -2.7126 | 2.6659 | |
| B - G | 1.0000 | -0.9016 | 2.9016 | |
| B - F | 1.2167 | -0.5621 | 2.9954 | |
| B - C | 1.4407 | -0.2602 | 3.1415 | |
| B - A | 1.7974 | 0.3157 | 3.2791 | *** |
| B - D | 1.9100 | 0.0084 | 3.8116 | *** |
| B - I | 2.2067 | 0.4279 | 3.9854 | *** |
| B - H | 2.5767 | 0.4506 | 4.7027 | *** |
| B - J | 2.5767 | 0.4506 | 4.7027 | *** |
| B - E | 2.5767 | -0.1126 | 5.2659 | |
| G - K | -1.0233 | -3.7126 | 1.6659 | |
| G - B | -1.0000 | -2.9016 | 0.9016 | |
| G - F | 0.2167 | -1.5621 | 1.9954 | |
| G - C | 0.4407 | -1.2602 | 2.1415 | |
| G - A | 0.7974 | -0.6843 | 2.2791 | |
| G - D | 0.9100 | -0.9916 | 2.8116 | |
| G - I | 1.2067 | -0.5721 | 2.9854 | |
| G - H | 1.5767 | -0.5494 | 3.7027 | |
| G - J | 1.5767 | -0.5494 | 3.7027 | |
| G - E | 1.5767 | -1.1126 | 4.2659 | |
| F - K | -1.2400 | -3.8439 | 1.3639 | |
| F - B | -1.2167 | -2.9954 | 0.5621 | |
| F - G | -0.2167 | -1.9954 | 1.5621 | |

The GLM Procedure

t Tests (LSD) for Lcfu

Comparisons significant at the 0.05 level are indicated by ***.

| Brand Comparison | Difference Between Means | 95% Confidence Limits | | |
|------------------|--------------------------|-----------------------|---------|-----|
| F - C | 0.2240 | -1.3383 | 1.7863 | |
| F - A | 0.5807 | -0.7397 | 1.9011 | |
| F - D | 0.6933 | -1.0854 | 2.4721 | |
| F - I | 0.9900 | -0.6568 | 2.6368 | |
| F - H | 1.3600 | -0.6569 | 3.3769 | |
| F - J | 1.3600 | -0.6569 | 3.3769 | |
| F - E | 1.3600 | -1.2439 | 3.9639 | |
| C - K | -1.4640 | -4.0152 | 1.0872 | |
| C - B | -1.4407 | -3.1415 | 0.2602 | |
| C - G | -0.4407 | -2.1415 | 1.2602 | |
| C - F | -0.2240 | -1.7863 | 1.3383 | |
| C - A | 0.3567 | -0.8566 | 1.5701 | |
| C - D | 0.4693 | -1.2315 | 2.1702 | |
| C - I | 0.7660 | -0.7963 | 2.3283 | |
| C - H | 1.1360 | -0.8125 | 3.0845 | |
| C - J | 1.1360 | -0.8125 | 3.0845 | |
| C - E | 1.1360 | -1.4152 | 3.6872 | |
| A - K | -1.8207 | -4.2314 | 0.5900 | |
| A - B | -1.7974 | -3.2791 | -0.3157 | *** |
| A - G | -0.7974 | -2.2791 | 0.6843 | |
| A - F | -0.5807 | -1.9011 | 0.7397 | |
| A - C | -0.3567 | -1.5701 | 0.8566 | |
| A - D | 0.1126 | -1.3691 | 1.5943 | |
| A - I | 0.4093 | -0.9111 | 1.7297 | |
| A - H | 0.7793 | -0.9812 | 2.5398 | |
| A - J | 0.7793 | -0.9812 | 2.5398 | |
| A - E | 0.7793 | -1.6314 | 3.1900 | |
| D - K | -1.9333 | -4.6226 | 0.7559 | |
| D - B | -1.9100 | -3.8116 | -0.0084 | *** |
| D - G | -0.9100 | -2.8116 | 0.9916 | |
| D - F | -0.6933 | -2.4721 | 1.0854 | |
| D - C | -0.4693 | -2.1702 | 1.2315 | |
| D - A | -0.1126 | -1.5943 | 1.3691 | |
| D - I | 0.2967 | -1.4821 | 2.0754 | |
| D - H | 0.6667 | -1.4594 | 2.7927 | |
| D - J | 0.6667 | -1.4594 | 2.7927 | |
| D - E | 0.6667 | -2.0226 | 3.3559 | |
| I - K | -2.2300 | -4.8339 | 0.3739 | |
| I - B | -2.2067 | -3.9854 | -0.4279 | *** |
| I - G | -1.2067 | -2.9854 | 0.5721 | |
| I - F | -0.9900 | -2.6368 | 0.6568 | |
| I - C | -0.7660 | -2.3283 | 0.7963 | |

The GLM Procedure

t Tests (LSD) for Lcfu

Comparisons significant at the 0.05 level are indicated by ***.

| Brand Comparison | Difference Between Means | 95% Confidence Limits | | |
|------------------|--------------------------|-----------------------|---------|-----|
| I - A | -0.4093 | -1.7297 | 0.9111 | |
| I - D | -0.2967 | -2.0754 | 1.4821 | |
| I - H | 0.3700 | -1.6469 | 2.3869 | |
| I - J | 0.3700 | -1.6469 | 2.3869 | |
| I - E | 0.3700 | -2.2339 | 2.9739 | |
| H - K | -2.6000 | -5.4524 | 0.2524 | |
| H - B | -2.5767 | -4.7027 | -0.4506 | *** |
| H - G | -1.5767 | -3.7027 | 0.5494 | |
| H - F | -1.3600 | -3.3769 | 0.6569 | |
| H - C | -1.1360 | -3.0845 | 0.8125 | |
| H - A | -0.7793 | -2.5398 | 0.9812 | |
| H - D | -0.6667 | -2.7927 | 1.4594 | |
| H - I | -0.3700 | -2.3869 | 1.6469 | |
| H - J | 0.0000 | -2.3290 | 2.3290 | |
| H - E | 0.0000 | -2.8524 | 2.8524 | |
| J - K | -2.6000 | -5.4524 | 0.2524 | |
| J - B | -2.5767 | -4.7027 | -0.4506 | *** |
| J - G | -1.5767 | -3.7027 | 0.5494 | |
| J - F | -1.3600 | -3.3769 | 0.6569 | |
| J - C | -1.1360 | -3.0845 | 0.8125 | |
| J - A | -0.7793 | -2.5398 | 0.9812 | |
| J - D | -0.6667 | -2.7927 | 1.4594 | |
| J - I | -0.3700 | -2.3869 | 1.6469 | |
| J - H | 0.0000 | -2.3290 | 2.3290 | |
| J - E | 0.0000 | -2.8524 | 2.8524 | |
| E - K | -2.6000 | -5.8936 | 0.6936 | |
| E - B | -2.5767 | -5.2659 | 0.1126 | |
| E - G | -1.5767 | -4.2659 | 1.1126 | |
| E - F | -1.3600 | -3.9639 | 1.2439 | |
| E - C | -1.1360 | -3.6872 | 1.4152 | |
| E - A | -0.7793 | -3.1900 | 1.6314 | |
| E - D | -0.6667 | -3.3559 | 2.0226 | |
| E - I | -0.3700 | -2.9739 | 2.2339 | |
| E - H | 0.0000 | -2.8524 | 2.8524 | |
| E - J | 0.0000 | -2.8524 | 2.8524 | |

The GLM Procedure

t Tests (LSD) for Lcfu

NOTE: This test controls the Type I comparisonwise error rate, not the experimentwise error rate.

| | |
|--------------------------|---------|
| Alpha | 0.05 |
| Error Degrees of Freedom | 25 |
| Error Mean Square | 1.27874 |
| Critical Value of t | 2.05954 |

Comparisons significant at the 0.05 level are indicated by ***.

| Flavor Comparison | Difference Between Means | 95% Confidence Limits | |
|----------------------|--------------------------------|--------------------------|--------|
| HOT - ORI | 0.1028 | -1.3496 | 1.5551 |
| HOT - PEP | 0.3700 | -1.2067 | 1.9467 |
| HOT - SMO | 0.7050 | -1.0738 | 2.4838 |
| HOT - TER | 0.7111 | -0.8415 | 2.2637 |
| ORI - HOT | -0.1028 | -1.5551 | 1.3496 |
| ORI - PEP | 0.2672 | -0.7224 | 1.2568 |
| ORI - SMO | 0.6022 | -0.6852 | 1.8896 |
| ORI - TER | 0.6083 | -0.3425 | 1.5591 |
| PEP - HOT | -0.3700 | -1.9467 | 1.2067 |
| PEP - ORI | -0.2672 | -1.2568 | 0.7224 |
| PEP - SMO | 0.3350 | -1.0912 | 1.7612 |
| PEP - TER | 0.3411 | -0.7906 | 1.4728 |
| SMO - HOT | -0.7050 | -2.4838 | 1.0738 |
| SMO - ORI | -0.6022 | -1.8896 | 0.6852 |
| SMO - PEP | -0.3350 | -1.7612 | 1.0912 |
| SMO - TER | 0.0061 | -1.3934 | 1.4056 |
| TER - HOT | -0.7111 | -2.2637 | 0.8415 |
| TER - ORI | -0.6083 | -1.5591 | 0.3425 |
| TER - PEP | -0.3411 | -1.4728 | 0.7906 |
| TER - SMO | -0.0061 | -1.4056 | 1.3934 |

The GLM Procedure

Class Level Information

| Class | Levels | Values |
|--------|--------|-----------------------|
| Brand | 11 | A B C D E F G H I J K |
| Flavor | 5 | HOT ORI PEP SMO TER |

| | |
|-----------------------------|----|
| Number of Observations Read | 42 |
| Number of Observations Used | 42 |

The GLM Procedure

Dependent Variable: Lcfu

| Source | DF | Sum of Squares | Mean Square | F Value | Pr > F |
|-----------------|----|----------------|-------------|---------|--------|
| Model | 16 | 21.00170787 | 1.31260674 | 1.03 | 0.4640 |
| Error | 25 | 31.96850403 | 1.27874016 | | |
| Corrected Total | 41 | 52.97021190 | | | |

| R-Square | Coeff Var | Root MSE | Lcfu Mean |
|----------|-----------|----------|-----------|
| 0.396481 | 57.51990 | 1.130814 | 1.965952 |

| Source | DF | Type I SS | Mean Square | F Value | Pr > F |
|--------|----|-------------|-------------|---------|--------|
| pH | 1 | 11.27211549 | 11.27211549 | 8.82 | 0.0065 |
| Aw | 1 | 0.00134174 | 0.00134174 | 0.00 | 0.9744 |
| Brand | 10 | 8.51528041 | 0.85152804 | 0.67 | 0.7445 |
| Flavor | 4 | 1.21297023 | 0.30324256 | 0.24 | 0.9147 |

| Source | DF | Type III SS | Mean Square | F Value | Pr > F |
|--------|----|-------------|-------------|---------|--------|
| pH | 1 | 0.35184819 | 0.35184819 | 0.28 | 0.6045 |
| Aw | 1 | 0.71610695 | 0.71610695 | 0.56 | 0.4612 |
| Brand | 10 | 8.56584724 | 0.85658472 | 0.67 | 0.7411 |
| Flavor | 4 | 1.21297023 | 0.30324256 | 0.24 | 0.9147 |

The GLM Procedure

Class Level Information

| Class | Levels | Values |
|--------|--------|-----------------------|
| Brand | 11 | A B C D E F G H I J K |
| Flavor | 5 | HOT ORI PEP SMO TER |

| | |
|-----------------------------|----|
| Number of Observations Read | 42 |
| Number of Observations Used | 42 |

The GLM Procedure

Dependent Variable: Lcfu

| Source | DF | Sum of Squares | Mean Square | F Value | Pr > F |
|-----------------|----|----------------|-------------|---------|--------|
| Model | 16 | 21.00170787 | 1.31260674 | 1.03 | 0.4640 |
| Error | 25 | 31.96850403 | 1.27874016 | | |
| Corrected Total | 41 | 52.97021190 | | | |

| R-Square | Coeff Var | Root MSE | Lcfu Mean |
|----------|-----------|----------|-----------|
| 0.396481 | 57.51990 | 1.130814 | 1.965952 |

| Source | DF | Type I SS | Mean Square | F Value | Pr > F |
|--------|----|-------------|-------------|---------|--------|
| pH | 1 | 11.27211549 | 11.27211549 | 8.82 | 0.0065 |
| Aw | 1 | 0.00134174 | 0.00134174 | 0.00 | 0.9744 |
| Brand | 10 | 8.51528041 | 0.85152804 | 0.67 | 0.7445 |
| Flavor | 4 | 1.21297023 | 0.30324256 | 0.24 | 0.9147 |

| Source | DF | Type III SS | Mean Square | F Value | Pr > F |
|--------|----|-------------|-------------|---------|--------|
| pH | 1 | 0.35184819 | 0.35184819 | 0.28 | 0.6045 |
| Aw | 1 | 0.71610695 | 0.71610695 | 0.56 | 0.4612 |
| Brand | 10 | 8.56584724 | 0.85658472 | 0.67 | 0.7411 |
| Flavor | 4 | 1.21297023 | 0.30324256 | 0.24 | 0.9147 |

The GLM Procedure
Least Squares Means

| Brand | Lcfu LSMEAN | LSMEAN Number |
|-------|-------------|------------------|
| A | 1.97833858 | 1 |
| B | 3.19268902 | 2 |
| C | 2.05188315 | 3 |
| D | 1.72523665 | 4 |
| E | 0.96773691 | 5 |
| F | 1.66591361 | 6 |
| G | 2.89063024 | 7 |
| H | 1.22159104 | 8 |
| I | 1.21802088 | 9 |
| J | 1.08502324 | 10 |
| K | 3.13875263 | 11 |

Least Squares Means for Effect Brand
t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 1 | 2 | 3 | 4 | 5 | 6 |
|-----|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|
| 1 | | -1.19144 0.2447 | -0.10699 0.9156 | 0.318182 0.7530 | 0.782996 0.4410 | 0.302568 0.7647 |
| 2 | 1.191442 0.2447 | | 1.239374 0.2267 | 1.195257 0.2432 | 1.537316 0.1368 | 1.320874 0.1985 |
| 3 | 0.106993 0.9156 | -1.23937 0.2267 | | 0.348739 0.7302 | 0.825236 0.4170 | 0.368075 0.7159 |
| 4 | -0.31818 0.7530 | -1.19526 0.2432 | -0.34874 0.7302 | | 0.546631 0.5895 | 0.051277 0.9595 |
| 5 | -0.783 0.4410 | -1.53732 0.1368 | -0.82524 0.4170 | -0.54663 0.5895 | | -0.45562 0.6526 |
| 6 | -0.30257 0.7647 | -1.32087 0.1985 | -0.36807 0.7159 | -0.05128 0.9595 | 0.45562 0.6526 | |
| 7 | 1.083254 0.2890 | -0.26423 0.7938 | 0.913904 0.3695 | 1.0561 0.3010 | 1.358001 0.1866 | 0.889351 0.3823 |
| 8 | -0.82948 0.4147 | -1.62368 0.1170 | -0.83642 0.4108 | -0.43809 0.6651 | 0.168265 0.8677 | -0.32722 0.7462 |
| 9 | -0.93918 0.3566 | -1.67167 0.1071 | -0.91085 0.3711 | -0.56788 0.5752 | 0.186362 0.8537 | -0.42402 0.6752 |
| 10 | -0.9256 0.3635 | -1.48075 0.1512 | -0.85894 0.3985 | -0.60403 0.5513 | 0.075502 0.9404 | -0.45664 0.6519 |
| 11 | 0.840923 0.4084 | -0.04087 0.9677 | 0.82866 0.4151 | 0.915133 0.3689 | 1.231845 0.2295 | 1.016783 0.3190 |

The GLM Procedure
Least Squares Means

Least Squares Means for Effect Brand
t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 7 | 8 | 9 | 10 | 11 |
|-----|--------------------|--------------------|--------------------|--------------------|--------------------|
| 1 | -1.08325 0.2890 | 0.829476 0.4147 | 0.939178 0.3566 | 0.925603 0.3635 | -0.84092 0.4084 |
| 2 | 0.264229 0.7938 | 1.623677 0.1170 | 1.671669 0.1071 | 1.480753 0.1512 | 0.040873 0.9677 |
| 3 | -0.9139 0.3695 | 0.836421 0.4108 | 0.910852 0.3711 | 0.858936 0.3985 | -0.82866 0.4151 |
| 4 | -1.0561 0.3010 | 0.438094 0.6651 | 0.567882 0.5752 | 0.604033 0.5513 | -0.91513 0.3689 |
| 5 | -1.358 0.1866 | -0.16826 0.8677 | -0.18636 0.8537 | -0.0755 0.9404 | -1.23185 0.2295 |
| 6 | -0.88935 0.3823 | 0.327221 0.7462 | 0.424021 0.6752 | 0.456637 0.6519 | -1.01678 0.3190 |
| 7 | | 1.577912 0.1272 | 1.462387 0.1561 | 1.402898 0.1729 | -0.16517 0.8701 |
| 8 | -1.57791 0.1272 | | 0.003007 0.9976 | 0.105695 0.9167 | -1.25117 0.2225 |
| 9 | -1.46239 0.1561 | -0.00301 0.9976 | | 0.127269 0.8997 | -1.27419 0.2143 |
| 10 | -1.4029 0.1729 | -0.1057 0.9167 | -0.12727 0.8997 | | -1.21978 0.2339 |
| 11 | 0.165168 0.8701 | 1.251169 0.2225 | 1.274191 0.2143 | 1.219784 0.2339 | |

NOTE: To ensure overall protection level, only probabilities associated with pre-planned comparisons should be used.

| Flavor | Lcfu LSMEAN | LSMEAN Number |
|--------|-------------|------------------|
| HOT | 2.30184536 | 1 |
| ORI | 2.08654162 | 2 |
| PEP | 1.96851211 | 3 |
| SMO | 1.56617668 | 4 |
| TER | 1.68411329 | 5 |

The GLM Procedure
Least Squares Means

Least Squares Means for Effect Flavor
t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 1 | 2 | 3 | 4 | 5 |
|-----|--------------------|--------------------|--------------------|--------------------|--------------------|
| 1 | | 0.257254 0.7991 | 0.344482 0.7334 | 0.707389 0.4859 | 0.656547 0.5175 |
| 2 | -0.25725 0.7991 | | 0.218099 0.8291 | 0.708631 0.4851 | 0.765869 0.4509 |
| 3 | -0.34448 0.7334 | -0.2181 0.8291 | | 0.480676 0.6349 | 0.439222 0.6643 |
| 4 | -0.70739 0.4859 | -0.70863 0.4851 | -0.48068 0.6349 | | -0.16129 0.8732 |
| 5 | -0.65655 0.5175 | -0.76587 0.4509 | -0.43922 0.6643 | 0.161292 0.8732 | |

NOTE: To ensure overall protection level, only probabilities associated with pre-planned comparisons should be used.

The GLM Procedure

Class Level Information

| Class | Levels | Values |
|--------|--------|---------------------------------------|
| pH | 10 | 5.1 5.3 5.4 5.5 5.6 5.7 5.8 5.9 6 6.1 |
| Brand | 11 | A B C D E F G H I J K |
| Flavor | 5 | HOT ORI PEP SMO TER |

Number of Observations Read 42
Number of Observations Used 42

The GLM Procedure

Dependent Variable: Lcfu

| Source | DF | Sum of Squares | Mean Square | F Value |
|-----------------|----|----------------|-------------|---------|
| Model | 24 | 31.97260414 | 1.33219184 | 1.08 |
| Error | 17 | 20.99760776 | 1.23515340 | |
| Corrected Total | 41 | 52.97021190 | | |

| Source | Pr > F |
|-----------------|--------|
| Model | 0.4441 |
| Error | |
| Corrected Total | |

| R-Square | Coeff Var | Root MSE | Lcfu Mean |
|----------|-----------|----------|-----------|
| 0.603596 | 56.53110 | 1.111375 | 1.965952 |

| Source | DF | Type I SS | Mean Square | F Value |
|--------|----|-------------|-------------|---------|
| pH | 9 | 19.19203440 | 2.13244827 | 1.73 |
| Aw | 1 | 0.02934988 | 0.02934988 | 0.02 |
| Brand | 10 | 12.26243364 | 1.22624336 | 0.99 |
| Flavor | 4 | 0.48878622 | 0.12219656 | 0.10 |

| Source | Pr > F |
|--------|--------|
| pH | 0.1588 |
| Aw | 0.8793 |
| Brand | 0.4854 |
| Flavor | 0.9814 |

| Source | DF | Type III SS | Mean Square | F Value |
|--------|----|-------------|-------------|---------|
| pH | 9 | 11.32274445 | 1.25808272 | 1.02 |
| Aw | 1 | 0.65088018 | 0.65088018 | 0.53 |

| Source | Pr > F |
|--------|--------|
| pH | 0.4636 |
| Aw | 0.4778 |

The GLM Procedure

Dependent Variable: Lcfu

| Source | DF | Type III SS | Mean Square | F Value |
|--------|----|-------------|-------------|---------|
| Brand | 10 | 11.63150609 | 1.16315061 | 0.94 |
| Flavor | 4 | 0.48878622 | 0.12219656 | 0.10 |

| Source | Pr > F |
|--------|--------|
| Brand | 0.5220 |
| Flavor | 0.9814 |

The GLM Procedure
 Least Squares Means

| pH | Lcfu LSMEAN | LSMEAN Number |
|-----|-------------|---------------|
| 5.1 | 0.97290460 | 1 |
| 5.3 | 2.26350206 | 2 |
| 5.4 | 2.75100855 | 3 |
| 5.5 | 1.71226730 | 4 |
| 5.6 | 1.97140847 | 5 |
| 5.7 | 3.96596228 | 6 |
| 5.8 | 1.27695223 | 7 |
| 5.9 | 1.17763858 | 8 |
| 6 | 1.51605047 | 9 |
| 6.1 | 3.72326613 | 10 |

Least Squares Means for Effect pH
 t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 1 | 2 | 3 | 4 | 5 |
|-----|--------------------|--------------------|--------------------|--------------------|--------------------|
| 1 | | -0.80621 0.4313 | -0.75742 0.4592 | -0.42117 0.6789 | -0.49463 0.6272 |
| 2 | 0.806206 0.4313 | | -0.1982 0.8452 | 0.308438 0.7615 | 0.138813 0.8912 |
| 3 | 0.757421 0.4592 | 0.1982 0.8452 | | 0.738072 0.4705 | 0.698615 0.4942 |
| 4 | 0.421172 0.6789 | -0.30844 0.7615 | -0.73807 0.4705 | | -0.26996 0.7904 |
| 5 | 0.494627 0.6272 | -0.13881 0.8912 | -0.69861 0.4942 | 0.269955 0.7904 | |
| 6 | 1.342105 0.1972 | 0.746559 0.4655 | 0.775033 0.4490 | 1.571707 0.1344 | 1.470711 0.1596 |
| 7 | 0.143993 0.8872 | -0.44752 0.6601 | -1.43008 0.1708 | -0.42528 0.6760 | -1.03805 0.3138 |
| 8 | 0.102238 0.9198 | -0.5244 0.6068 | -1.36065 0.1914 | -0.57207 0.5748 | -1.12049 0.2781 |
| 9 | 0.27454 0.7870 | -0.36682 0.7183 | -0.76255 0.4562 | -0.1526 0.8805 | -0.387 0.7036 |
| 10 | 0.948714 0.3561 | 0.490828 0.6298 | 0.53825 0.5974 | 1.020267 0.3219 | 0.999091 0.3318 |

The GLM Procedure
 Least Squares Means

Least Squares Means for Effect pH
 t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 6 | 7 | 8 | 9 | 10 |
|-----|--------------------|--------------------|--------------------|--------------------|--------------------|
| 1 | -1.3421 0.1972 | -0.14399 0.8872 | -0.10224 0.9198 | -0.27454 0.7870 | -0.94871 0.3561 |
| 2 | -0.74656 0.4655 | 0.447525 0.6601 | 0.524397 0.6068 | 0.366823 0.7183 | -0.49083 0.6298 |
| 3 | -0.77503 0.4490 | 1.430082 0.1708 | 1.360651 0.1914 | 0.762546 0.4562 | -0.53825 0.5974 |
| 4 | -1.57171 0.1344 | 0.425283 0.6760 | 0.572065 0.5748 | 0.152602 0.8805 | -1.02027 0.3219 |
| 5 | -1.47071 0.1596 | 1.038048 0.3138 | 1.120491 0.2781 | 0.387002 0.7036 | -0.99909 0.3318 |
| 6 | | 1.925633 0.0710 | 1.999627 0.0618 | 1.498328 0.1524 | 0.111232 0.9127 |
| 7 | -1.92563 0.0710 | | 0.149734 0.8827 | -0.187 0.8539 | -1.5715 0.1345 |
| 8 | -1.99963 0.0618 | -0.14973 0.8827 | | -0.301 0.7671 | -1.43689 0.1689 |
| 9 | -1.49833 0.1524 | 0.187003 0.8539 | 0.301002 0.7671 | | -0.97835 0.3416 |
| 10 | -0.11123 0.9127 | 1.571498 0.1345 | 1.436885 0.1689 | 0.97835 0.3416 | |

NOTE: To ensure overall protection level, only probabilities associated with pre-planned comparisons should be used.

| Brand | Lcfu LSMEAN | LSMEAN Number |
|-------|-------------|---------------|
| A | 2.60006847 | 1 |
| B | 4.11366803 | 2 |
| C | 1.98532729 | 3 |
| D | 1.66266920 | 4 |
| E | -0.53590746 | 5 |
| F | 1.98485302 | 6 |
| G | 3.38576102 | 7 |
| H | 1.66839701 | 8 |
| I | 1.86257863 | 9 |
| J | 1.37945146 | 10 |
| K | 3.35719006 | 11 |

The GLM Procedure
Least Squares Means

Least Squares Means for Effect Brand
t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 1 | 2 | 3 | 4 | 5 | 6 |
|-----|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|
| 1 | | -0.85197 0.4061 | 0.798275 0.4357 | 0.985122 0.3384 | 1.583626 0.1317 | 0.515522 0.6128 |
| 2 | 0.851974 0.4061 | | 1.272498 0.2203 | 1.243498 0.2306 | 2.179031 0.0437 | 1.179971 0.2543 |
| 3 | -0.79827 0.4357 | -1.2725 0.2203 | | 0.294553 0.7719 | 1.410733 0.1764 | 0.00039 0.9997 |
| 4 | -0.98512 0.3384 | -1.2435 0.2306 | -0.29455 0.7719 | | 1.011806 0.3258 | -0.23553 0.8166 |
| 5 | -1.58363 0.1317 | -2.17903 0.0437 | -1.41073 0.1764 | -1.01181 0.3258 | | -1.17123 0.2577 |
| 6 | -0.51552 0.6128 | -1.17997 0.2543 | -0.00039 0.9997 | 0.235533 0.8166 | 1.17123 0.2577 | |
| 7 | 0.84657 0.4090 | -0.35494 0.7270 | 1.312139 0.2069 | 1.337035 0.1988 | 1.817814 0.0868 | 0.829976 0.4181 |
| 8 | -0.98827 0.3369 | -1.17494 0.2562 | -0.2844 0.7795 | 0.004609 0.9964 | 0.991957 0.3351 | -0.20608 0.8392 |
| 9 | -0.62785 0.5384 | -1.46966 0.1599 | -0.10034 0.9212 | 0.134983 0.8942 | 1.277331 0.2187 | -0.0866 0.9320 |
| 10 | -0.8492 0.4076 | -1.47646 0.1581 | -0.40131 0.6932 | -0.16613 0.8700 | 0.908288 0.3764 | -0.33812 0.7394 |
| 11 | 0.308367 0.7615 | -0.47565 0.6404 | 0.579109 0.5701 | 0.655766 0.5208 | 1.450507 0.1651 | 0.57204 0.5748 |

Least Squares Means for Effect Brand
t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 7 | 8 | 9 | 10 | 11 |
|-----|--------------------|--------------------|--------------------|--------------------|--------------------|
| 1 | -0.84657 0.4090 | 0.988273 0.3369 | 0.627849 0.5384 | 0.849202 0.4076 | -0.30837 0.7615 |
| 2 | 0.354939 0.7270 | 1.174941 0.2562 | 1.46966 0.1599 | 1.476462 0.1581 | 0.475649 0.6404 |
| 3 | -1.31214 0.2069 | 0.284398 0.7795 | 0.100342 0.9212 | 0.401308 0.6932 | -0.57911 0.5701 |
| 4 | -1.33704 0.1988 | -0.00461 0.9964 | -0.13498 0.8942 | 0.166133 0.8700 | -0.65577 0.5208 |
| 5 | -1.81781 0.0868 | -0.99196 0.3351 | -1.27733 0.2187 | -0.90829 0.3764 | -1.45051 0.1651 |
| 6 | -0.82998 0.4181 | 0.206079 0.8392 | 0.086597 0.9320 | 0.338119 0.7394 | -0.57204 0.5748 |

The GLM Procedure
 Least Squares Means

Least Squares Means for Effect Brand
 t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 7 | 8 | 9 | 10 | 11 |
|-----|--------------------|--------------------|--------------------|--------------------|--------------------|
| 7 | | 1.517447 0.1475 | 0.964813 0.3482 | 1.138648 0.2706 | 0.010577 0.9917 |
| 8 | -1.51745 0.1475 | | -0.12151 0.9047 | 0.161981 0.8732 | -0.62332 0.5413 |
| 9 | -0.96481 0.3482 | 0.121509 0.9047 | | 0.434749 0.6692 | -0.6739 0.5094 |
| 10 | -1.13865 0.2706 | -0.16198 0.8732 | -0.43475 0.6692 | | -0.8033 0.4329 |
| 11 | -0.01058 0.9917 | 0.623322 0.5413 | 0.6739 0.5094 | 0.803296 0.4329 | |

NOTE: To ensure overall protection level, only probabilities associated with pre-planned comparisons should be used.

| Flavor | Lcfu LSMEAN | LSMEAN Number |
|--------|-------------|---------------|
| HOT | 2.48230421 | 1 |
| ORI | 2.23184398 | 2 |
| PEP | 1.86660077 | 3 |
| SMO | 1.91134346 | 4 |
| TER | 2.17338791 | 5 |

Least Squares Means for Effect Flavor
 t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 1 | 2 | 3 | 4 | 5 |
|-----|--------------------|--------------------|--------------------|--------------------|--------------------|
| 1 | | 0.250057 0.8055 | 0.43436 0.6695 | 0.479308 0.6378 | 0.279465 0.7833 |
| 2 | -0.25006 0.8055 | | 0.46419 0.6484 | 0.375016 0.7123 | 0.098709 0.9225 |
| 3 | -0.43436 0.6695 | -0.46419 0.6484 | | -0.04083 0.9679 | -0.34667 0.7331 |
| 4 | -0.47931 0.6378 | -0.37502 0.7123 | 0.040833 0.9679 | | -0.34742 0.7325 |

The GLM Procedure
Least Squares MeansLeast Squares Means for Effect Flavor
t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 1 | 2 | 3 | 4 | 5 |
|-----|--------------------|--------------------|--------------------|--------------------|---|
| 5 | -0.27946 0.7833 | -0.09871 0.9225 | 0.346671 0.7331 | 0.347422 0.7325 | |

NOTE: To ensure overall protection level, only probabilities associated with pre-planned comparisons should be used.

The REG Procedure
 Model: MODEL1
 Dependent Variable: Lcfu

Number of Observations Read 42
 Number of Observations Used 42

Analysis of Variance

| Source | DF | Sum of Squares | Mean Square | F Value |
|-----------------|----|----------------|-------------|---------|
| Model | 1 | 11.27212 | 11.27212 | 10.81 |
| Error | 40 | 41.69810 | 1.04245 | |
| Corrected Total | 41 | 52.97021 | | |

Analysis of Variance

| Source | Pr > F |
|-----------------|--------|
| Model | 0.0021 |
| Error | |
| Corrected Total | |

Root MSE 1.02101 R-Square 0.2128
 Dependent Mean 1.96595 Adj R-Sq 0.1931
 Coeff Var 51.93440

Parameter Estimates

| Variable | DF | Parameter Estimate | Standard Error | t Value | Pr > t |
|-----------|----|--------------------|----------------|---------|---------|
| Intercept | 1 | 15.27932 | 4.05174 | 3.77 | 0.0005 |
| pH | 1 | -2.32887 | 0.70822 | -3.29 | 0.0021 |

The GLM Procedure

Class Level Information

| Class | Levels | Values |
|--------|--------|-----------------------|
| Brand | 11 | A B C D E F G H I J K |
| Flavor | 5 | HOT ORI PEP SMO TER |

| | |
|-----------------------------|----|
| Number of Observations Read | 42 |
| Number of Observations Used | 42 |

The GLM Procedure

Dependent Variable: Lcfu

| Source | DF | Sum of Squares | Mean Square | F Value |
|-----------------|----|----------------|-------------|---------|
| Model | 16 | 21.00170787 | 1.31260674 | 1.03 |
| Error | 25 | 31.96850403 | 1.27874016 | |
| Corrected Total | 41 | 52.97021190 | | |

Source Pr > F

Model 0.4640

Error

Corrected Total

| R-Square | Coeff Var | Root MSE | Lcfu Mean |
|----------|-----------|----------|-----------|
| 0.396481 | 57.51990 | 1.130814 | 1.965952 |

| Source | DF | Type I SS | Mean Square | F Value |
|--------|----|-------------|-------------|---------|
| pH | 1 | 11.27211549 | 11.27211549 | 8.82 |
| Aw | 1 | 0.00134174 | 0.00134174 | 0.00 |
| Brand | 10 | 8.51528041 | 0.85152804 | 0.67 |
| Flavor | 4 | 1.21297023 | 0.30324256 | 0.24 |

Source Pr > F

pH 0.0065

Aw 0.9744

Brand 0.7445

Flavor 0.9147

| Source | DF | Type III SS | Mean Square | F Value |
|--------|----|-------------|-------------|---------|
| pH | 1 | 0.35184819 | 0.35184819 | 0.28 |
| Aw | 1 | 0.71610695 | 0.71610695 | 0.56 |

Source Pr > F

pH 0.6045

Aw 0.4612

The GLM Procedure

Dependent Variable: Lcfu

| Source | DF | Type III SS | Mean Square | F Value |
|--------|----|-------------|-------------|---------|
| Brand | 10 | 8.56584724 | 0.85658472 | 0.67 |
| Flavor | 4 | 1.21297023 | 0.30324256 | 0.24 |

| Source | Pr > F |
|--------|--------|
| Brand | 0.7411 |
| Flavor | 0.9147 |

The GLM Procedure
 Least Squares Means

| Brand | Lcfu LSMEAN | LSMEAN Number |
|-------|-------------|---------------|
| A | 1.97833858 | 1 |
| B | 3.19268902 | 2 |
| C | 2.05188315 | 3 |
| D | 1.72523665 | 4 |
| E | 0.96773691 | 5 |
| F | 1.66591361 | 6 |
| G | 2.89063024 | 7 |
| H | 1.22159104 | 8 |
| I | 1.21802088 | 9 |
| J | 1.08502324 | 10 |
| K | 3.13875263 | 11 |

Least Squares Means for Effect Brand
 t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 1 | 2 | 3 | 4 | 5 | 6 |
|-----|----------|----------|----------|----------|----------|----------|
| 1 | | -1.19144 | -0.10699 | 0.318182 | 0.782996 | 0.302568 |
| | | 0.2447 | 0.9156 | 0.7530 | 0.4410 | 0.7647 |
| 2 | 1.191442 | | 1.239374 | 1.195257 | 1.537316 | 1.320874 |
| | 0.2447 | | 0.2267 | 0.2432 | 0.1368 | 0.1985 |
| 3 | 0.106993 | -1.23937 | | 0.348739 | 0.825236 | 0.368075 |
| | 0.9156 | 0.2267 | | 0.7302 | 0.4170 | 0.7159 |
| 4 | -0.31818 | -1.19526 | -0.34874 | | 0.546631 | 0.051277 |
| | 0.7530 | 0.2432 | 0.7302 | | 0.5895 | 0.9595 |
| 5 | -0.783 | -1.53732 | -0.82524 | -0.54663 | | -0.45562 |
| | 0.4410 | 0.1368 | 0.4170 | 0.5895 | | 0.6526 |
| 6 | -0.30257 | -1.32087 | -0.36807 | -0.05128 | 0.45562 | |
| | 0.7647 | 0.1985 | 0.7159 | 0.9595 | 0.6526 | |
| 7 | 1.083254 | -0.26423 | 0.913904 | 1.0561 | 1.358001 | 0.889351 |
| | 0.2890 | 0.7938 | 0.3695 | 0.3010 | 0.1866 | 0.3823 |
| 8 | -0.82948 | -1.62368 | -0.83642 | -0.43809 | 0.168265 | -0.32722 |
| | 0.4147 | 0.1170 | 0.4108 | 0.6651 | 0.8677 | 0.7462 |
| 9 | -0.93918 | -1.67167 | -0.91085 | -0.56788 | 0.186362 | -0.42402 |
| | 0.3566 | 0.1071 | 0.3711 | 0.5752 | 0.8537 | 0.6752 |
| 10 | -0.9256 | -1.48075 | -0.85894 | -0.60403 | 0.075502 | -0.45664 |
| | 0.3635 | 0.1512 | 0.3985 | 0.5513 | 0.9404 | 0.6519 |
| 11 | 0.840923 | -0.04087 | 0.82866 | 0.915133 | 1.231845 | 1.016783 |
| | 0.4084 | 0.9677 | 0.4151 | 0.3689 | 0.2295 | 0.3190 |

The GLM Procedure
Least Squares Means

Least Squares Means for Effect Brand
t for H0: LSMEAN(i)=LSMEAN(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 7 | 8 | 9 | 10 | 11 |
|-----|--------------------|--------------------|--------------------|--------------------|--------------------|
| 1 | -1.08325 0.2890 | 0.829476 0.4147 | 0.939178 0.3566 | 0.925603 0.3635 | -0.84092 0.4084 |
| 2 | 0.264229 0.7938 | 1.623677 0.1170 | 1.671669 0.1071 | 1.480753 0.1512 | 0.040873 0.9677 |
| 3 | -0.9139 0.3695 | 0.836421 0.4108 | 0.910852 0.3711 | 0.858936 0.3985 | -0.82866 0.4151 |
| 4 | -1.0561 0.3010 | 0.438094 0.6651 | 0.567882 0.5752 | 0.604033 0.5513 | -0.91513 0.3689 |
| 5 | -1.358 0.1866 | -0.16826 0.8677 | -0.18636 0.8537 | -0.0755 0.9404 | -1.23185 0.2295 |
| 6 | -0.88935 0.3823 | 0.327221 0.7462 | 0.424021 0.6752 | 0.456637 0.6519 | -1.01678 0.3190 |
| 7 | | 1.577912 0.1272 | 1.462387 0.1561 | 1.402898 0.1729 | -0.16517 0.8701 |
| 8 | -1.57791 0.1272 | | 0.003007 0.9976 | 0.105695 0.9167 | -1.25117 0.2225 |
| 9 | -1.46239 0.1561 | -0.00301 0.9976 | | 0.127269 0.8997 | -1.27419 0.2143 |
| 10 | -1.4029 0.1729 | -0.1057 0.9167 | -0.12727 0.8997 | | -1.21978 0.2339 |
| 11 | 0.165168 0.8701 | 1.251169 0.2225 | 1.274191 0.2143 | 1.219784 0.2339 | |

NOTE: To ensure overall protection level, only probabilities associated with pre-planned comparisons should be used.

| Flavor | Lcfu LSMEAN | LSMEAN Number |
|--------|-------------|---------------|
| HOT | 2.30184536 | 1 |
| ORI | 2.08654162 | 2 |
| PEP | 1.96851211 | 3 |
| SMO | 1.56617668 | 4 |
| TER | 1.68411329 | 5 |

The GLM Procedure
Least Squares Means

Least Squares Means for Effect Flavor
t for H0: LSMean(i)=LSMean(j) / Pr > |t|

Dependent Variable: Lcfu

| i/j | 1 | 2 | 3 | 4 | 5 |
|-----|--------------------|--------------------|--------------------|--------------------|--------------------|
| 1 | | 0.257254 0.7991 | 0.344482 0.7334 | 0.707389 0.4859 | 0.656547 0.5175 |
| 2 | -0.25725 0.7991 | | 0.218099 0.8291 | 0.708631 0.4851 | 0.765869 0.4509 |
| 3 | -0.34448 0.7334 | -0.2181 0.8291 | | 0.480676 0.6349 | 0.439222 0.6643 |
| 4 | -0.70739 0.4859 | -0.70863 0.4851 | -0.48068 0.6349 | | -0.16129 0.8732 |
| 5 | -0.65655 0.5175 | -0.76587 0.4509 | -0.43922 0.6643 | 0.161292 0.8732 | |

NOTE: To ensure overall protection level, only probabilities associated with pre-planned comparisons should be used.

VITA

Miriam Velasco Ramos

Candidate for the Degree of

Master of Science

Thesis: SURVEY OF MICROBIOLOGICAL CONTENT OF
COMMERCIAL BEEF JERKY

Major Field: Food Sciences

Biographical:

Personal Data: Born in Durango, Dgo. México, on February, the daughter of Antonio Velasco Rincón y Leobarda Ramos López

Education: Graduated from CEBTIS 110 from Durango city High School, received Bachelor of Science degree in Agrochemistry Engineering from Juarez University at Durango State in February 1990. Completed the requirements for the Master of Science degree with a major in Food Science at Oklahoma State University (December, 2007).

Experience: Employed as a Teacher in ICAC High School in 1990; manager of own business from 1991 to 2000; employed by Agricultural Department of Durango State as an agricultural extensionist from 1998 to 2002; employed by Oklahoma State University, FAPC as a graduate research assistant, 2005 to present.

Professional Memberships: Institute of Food Technologists since 2003.

Name: Miriam Velasco Ramos

Date of Degree: December, 2007

Institution: Oklahoma State University

Location: Stillwater, Oklahoma

Title of Study: SURVEY OF MICROBIOLOGICAL CONTENT OF
COMMERCIAL BEEF JERKY

Pages in Study: 78

Candidate for the Degree of Master of Science

Major Field: Food Science

Scope and Method of Study: Forty-two samples of beef jerky were analyzed. They were aseptically ground and appropriate serial dilutions plated by a spread plate method in duplicate on Baird Parker agar for coagulase positive *Staphylococcus aureus*, and on acidified Potato Dextrose agar for yeast and molds; the pour plate method was used for total plate count on Plate Count Agar, and for coliforms on Violet Red Bile agar. Analyses for *Salmonella* and *E. coli* O157:H7 were done using Minividas equipment following enrichment in Buffered Peptone Water for the former and Tryptic Soy Broth and Mac Conkey broth for the latter.

Findings and Conclusions: No pathogenic bacteria were detected in any sample; the numbers on Plate Count Agar were low. While for a few samples an occasional mold colony appeared on the lowest dilution plated (1:100) most had none. Since we did not detect any pathogens, it is tempting to assume that this product is safe to consume.

ADVISER'S APPROVAL: Dr. Stanley E. Gilliland
