FOOD SAFETY ISSUES AND PHYSICAL PROPERTIES ASSOCIATED
WITH HOME-STYLE BEEF JERKY

by

RUTH ANN ROSE

(Under the Direction of Mark A. Harrison)

ABSTRACT

Home-style jerky has grown in popularity over the past years because it is easy to prepare, lightweight, low fat, and tasty. However, during the 1980's and 1990's, several foodborne outbreaks, associated with home-style and small scale jerky processors, led people to re-examine the safety of the jerky making process. This thesis, addressed food safety issues related to *Escherichia coli* O157:H7, *Listeria monocytogenes*, and *Salmonella* spp. on preparation of home-style beef jerky. Antimicrobial effects on of sugar and salt marinades were compared. Sodium chloride levels on the jerky process were investigated as the antimicrobial effect of a low and a regular salt level marinade were compared. Whether acid adapted cells have a higher survival rate than nonadapted cells was investigated. The type of marinade did not have an effect on physical properties. The survival of the acid-adapted and nonadapted cells were not significantly different.

INDEX WORDS: Beef jerky, acid adaptation, *E. coli* O157:H7, *L. monocytogenes*, *Salmonella*, reduced salt, marination
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B.S., The University of Georgia, 1995

A Thesis Submitted to the Graduate Faculty of The University of Georgia in Partial
Fulfillment of the Requirements for the Degree

MASTER OF SCIENCE

ATHENS, GEORGIA

2003
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August 2003
ACKNOWLEDGMENTS

I extend my gratitude to Dr. Mark Harrison, Dr. Judy Harrison, and Dr. Huang for their contribution to my M.S. I also thank Kortney Karnok for the numerous hours she spent helping me in preparing supplies and sampling the beef jerky. Dr. Glenn Ware was instrumental in analyzing my years of data and I thank him for his time and effort. Dr. Elaine D’sa provided guidance throughout the experiments and she was helpful on all levels. I would like to thank Jin Kyung Kim for her expertise on the graphs and formatting of my thesis. I would like to also thank Robin Salinsky for volunteering to assist in sampling. My husband, Mike Morrow, has been very supportive of me earning my Masters and working full time. I know he is as thrilled as I am that I have finally graduated after all these years. I would like to thank the many others who contributed to my degree.
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INTRODUCTION

According to the World Health Organization foodborne illness is the most widespread health problem in the world (8). The Centers for Disease Control and Prevention (CDC) estimate 76 million illnesses occur annually in the U.S. alone (5) with 7,000 deaths projected due to foodborne illnesses (8). Economic loss due to productivity loss, medical cost, and food recalls amounts to $6.9 billion in just the U.S. (8). The four major pathogens of concern, *Campylobacter*, *Escherichia coli O157:H7*, *Listeria monocytogenes*, and *Salmonella*, account annually for an estimated 14 million illnesses, 60,000 hospitalizations, and 1,800 deaths in the U.S. (2). Consumers who prepare food for their own consumption or for small-scale retail purposes often face food safety issues. This is especially true for processes which may not reliably reduce the level of foodborne pathogens to a desired level.

Dehydration used as a food preservative has been practiced for hundreds of years (1). When properly done, this method of preservation has worked well in reducing the number of both spoilage and pathogenic bacteria on food. However, occasionally problems can occur. Beef jerky is a popular dehydrated food, but it was not until the 1980's and 1990's that the drying process for jerky was questioned when several illness outbreaks and recalls occurred. Studies have found that dehydration alternatives can provide safer jerky by having the internal temperature of the meat reach 71.1°C before or after the drying step (6). Although this is safer, the question of consumer acceptability needed to be addressed. Harrison et al. (4) found that a safer alternate method where the
jerky strips were marinated and dried in a 60°C dehydrator and then placed in a 135°C oven for 10 min was as acceptable to consumers as the traditional method (drying at 60°C) of marination and dehydration. Due to outbreaks of foodborne illness in recent years linked to other pathogens such as *E. coli* O157:H7 and *L. monocytogenes*, the USDA meat and poultry hotline (3) suggests steaming or roasting meat to 71.1°C before drying. The U.S. Food Safety and Inspection Service (FSIS) recommends a 5-log reduction in *E. coli* O157:H7 on jerky to be considered safe (7). However, no instructions are provided as to the exact procedure to accomplish this. Another recent concern that has become a food safety issue is the possibility that pathogenic cells can become habituated or tolerant to acid which could enable them to survive in the acidic conditions encountered in the marinated food and the host body. Since marinades for jerky are acidic, this potential food safety hazard requires further investigation. This thesis addresses various food safety issues associated with preparation of home-style beef jerky. Before decisions can be made with confidence concerning the inactivation of foodborne pathogens subjected to processing techniques and preservation agents, it is critical to acquire data that is relevant to real life situations. This is especially important when one considers the critical control points that would be evaluated for a minimally processed food operation. Such an example is prepared home-style beef jerky.
REFERENCES


Physical methods of food preservation use physical treatments to inhibit, destroy or remove undesirable microorganisms without introducing additives (Farkas, 1997). One way to accomplish this is to physically dehydrate the food. Dehydration is the oldest food preservation method and it works on the principle of limiting available water to unwanted microbes. Dehydrating reduces the water activity ($a_w$) which in turn reduces microbial growth and increases lag phase of bacteria resulting in biological forces being controlled (Farkas, 1997). As the temperature increases, the humidity decreases and the osmotic pressure increases. In 1795, the first dehydration room was invented (Desrosier, 1970) and over time people began to realize that all enzyme activity is arrested by dehydration (Rector, 1925). Dried foods are popular because they are nutritious, tasty and lightweight in addition to easy preparation and storage. Dried foods are high in fiber and low in fat which also adds to its appeal. Besides fruits and vegetables, meat has always been a popular food to dehydrate.

Bacteria can be introduced to red meat after removal of the hide through several routes (Jackson et al., 1997). The initial incision can introduce unwanted bacteria from the hide to the muscle. Contamination of workers’ hands, aerosols and dust from hide removal, the evisceration process, and fabrication are other ways to introduce unwanted microflora onto red meats. Bacterial attachment to muscle surface involves two stages. The first one involves a loose, reversible sorption believed to involve van der Waals forces. The second stage is an irreversible attachment involving the production of glycocalyx, an extracellular polysaccharide layer. Surface type, growth phase, temperature, cell density and motility also play a role in attachment.
PREVIOUS BEEF JERKY STUDIES

Holley (1995, 1998) examined the effectiveness of drying procedures in eliminating the risk of foodborne illness from *Salmonella*, *Staphylococcus*, *Bacillus subtilis* and *Clostridium perfringens*. Smith et al. (1977) investigated the effect of various drying methods on *Salmonella* and *Staphylococcus*. Both Holly and Smith and co-workers determined that heated drying, higher than 53.9°C, was necessary to reduce the risk of foodborne illness. In a more recent study, a reduction of 5.5-6.0 log CFU/g of *E. coli* O157:H7, *L. monocytogenes*, and *S. Typhimurium* was noted after cooking whole meat beef jerky strips prior to drying samples for 10 h (Harrison and Harrison, 1996). In uncooked samples, however, populations of the three organisms were reduced by only 3.3, 1.8 and 3.1 log, respectively, after 3 h of drying. In a separate study using ground beef jerky that either contained or lacked a nitrite cure mix, *E. coli* O157:H7, *L. monocytogenes*, and *S. Typhimurium* populations were only reduced by 2.5-4.0 logs CFU/g after 8 h of drying in samples that lacked the cure mix (Harrison et al., 1997, 1998). When cure mix was added, the bacterial populations were reduced by at least 4 logs. Addition of the cure mix resulted in a greater reduction in *E. coli* O157:H7 and *S. Typhimurium* populations. Faith et al. (1998) and Buege and Luchansky (1999) have shown in previous jerky experiments that the fat content of the meat can influence the reduction of pathogens. Meat with higher percentages of fat provided extra protection for the pathogens that were present. A correlation between drying temperature and time also exists. The higher the temperature, generally the less time required for complete
dehydration. However, care should be taken to not use temperatures that are too high, or case hardening will result in a strip that is not fully dehydrated.

In evaluating the efficacy of traditional preservation procedures, it is critical to consider that bacterial cells injured by such processes as heating, freezing and drying may undergo repair when conditions become more favorable or suitable for growth (e.g., during a storage period). This potential to regain the ability to cause illness is a particular threat with pathogens such as *E. coli* O157:H7 that are believed to have a low infectious dose (Griffin, 1991). There is evidence that when exposed to nonlethal stresses, many foodborne pathogens can undergo changes that may provide them with some degree of resistance to conditions that are typically considered lethal. For example, *E. coli* O157:H7, *L. monocytogenes* and *S. Typhimurium* can become more acid resistant and possibly more resistant to other stresses (e.g., heat, osmotic pressure), if subjected to relatively mild acidity before exposure to more acidic conditions (Buchanan and Doyle, 1997; Leyer et al., 1995; O’Driscoll et al., 1996; Ravishanker and Harrison, 1999). Growth of *E. coli* in an acidogenic (TSB with 1.0 % glucose) broth produced cells that expressed an acid resistance response while cells grown in a non-acidogenic (TSB without glucose) broth did not undergo the response (Buchanan and Edelson, 1999). Acid resistance was increased in those grown in the acidogenic broth. Most marinades used in jerky preparation are acidic in nature. Since foodborne pathogens may be present on the raw product which is exposed to the marinade, there is the possibility that the
microorganisms could undergo an adaptation that might become more resistant to the environmental stresses that the muscle tissue would receive during jerky processing.

Recovery of *E. coli* O157:H7 on two media types was examined by Harrison et al. (1998). This study examined the efficacy of various jerky preparation methods in reducing *E. coli* O157:H7 populations in ground beef and compared the recovery rate of *E. coli* O157:H7 on two selective plating media, modified sorbitol MacConkey agar and modified eosin methylene blue agar (MEMB). A trend toward slightly higher recovery rates with MEMB was observed.

*Effect of pre-treatments on E. coli O157:H7 inoculated onto whole beef jerky strips.*

A study by Albright et al. (2003) investigated the effect of 4 different pre-treatments of whole strip jerky on *E. coli* O157:H7. After 10 h of drying, the treatment marinated for 24 h at 4°C followed by submersion in pickle brine at 78°C for 90 s was shown to have the largest overall reduction and the highest pre-pretreatment reduction (5.7-5.8 log CFU/cm²). The other pre-treatments consisting of: (1) boiling at 94°C for 15 s plus marination at 4°C for 24 h; (2) immersing strips in a 1:1 vinegar and water solution for 20 s at 57.5°C plus marination at 4°C for 24 h and; (3) marination at 4°C for 24 h and then immersing in a 1:1 vinegar and water solution for 20 s at 57.5°C resulted in a 4.3- 4.5, 4.9-5.2 and 4.7- 4.8 log CFU/ cm² reduction, respectively (Albright et al., 2003). This research would be more valuable if sensory testing of the various pre-treatments was conducted so that the application of the safest method could be applied by the consumer.
Consumer acceptability and pathogen inactivation

Producing safe jerky that retains acceptable quality attributes is important. Lethality of *E. coli* O157:H7, *L. monocytogenes* and *Salmonella* as well as consumer acceptability and sensory attributes of jerky prepared by four methods (traditional, boil strips prior to drying, pre-cook strips to 71.1°C in an oven heated to 163.0°C prior to drying, and heating strips to 71.1°C oven after drying) were examined by Harrison et al. (2001). Of the four treatments, consumers preferred heating strips in the oven after drying even over the traditional method. The authors found that although the four treatments were significantly different in color, saltiness and texture, only texture appeared to influence overall consumer acceptability. Microbial challenge studies subjecting *E. coli* O157:H7, *L. monocytogenes* and *Salmonella* spp. to the four treatments resulted in log population reductions of 5.8, 3.9 and 4.6, respectively. Oven treatment of strips after drying reduced the pathogen populations by an additional 2 logs. A safer, acceptable home-dried beef jerky can be produced by oven-heating jerky strips after drying.

**Effect of various marinade treatments on acid and nonadapted** *L. monocytogenes*.

Calicioglu et al. (2003a) tested 5 various marinade treatments on whole strip jerky. All strips were dried at 60°C for 10 h. Results showed that dipping the strips into 1% Tween 20 for 15 min. and then into 5% acetic acid for 10 min. followed by traditional marination had the highest inactivation of *L. monocytogenes* (5.9-6.31 log CFU/cm²) and, the traditional marination had the least effect aside from the control strips which contained no marinade. The authors concluded that acid-adapted cells may not increase
resistance of microbes in jerky processing; thus their findings suggest no significant
differences between acid-adapted and nonadapted cells. The practicality of using Tween
20 in a home-prepared marinade may be questionable.

*Effect of various marinade treatments on acid-adapted and nonadapted* *L.*
*monocytogenes* *and E. coli O157:H7 inoculated onto beef jerky strips after drying.*

Calicioglu et al. (2002) used the same marinade treatments as they did in the
previous study. This time however, the jerky strips were inoculated after drying to
observe survival of *L. monocytogenes* during storage. The strips were stored at 25°C for
60 d and sampled at various intervals. By the end of the storage period, there was no
significant difference between acid-adapted and nonadapted cells. This same study was
done by Calicioglu et al. (2002) using *E. coli* O157:H7. This time, populations decreased
faster on jerky inoculated with acid-adapted rather than with nonadapted cells in all
marinade treatments. Rather than acid-adaption enhancing survival, the acid adaptation
*E. coli* O157:H7 enhanced inactivation during storage.

**ESCHERICHIA COLI**

*Types of Pathogenic*  *E. coli*

There are six types of pathogenic *E. coli* O157:H7. Enteroinvasive (EIEC) *E. coli*
causes nonbloody diarrhea, local tissue damage and is uncommon in the United States.
Enteroaggregative *E. coli* (EAggEC) effects children of developing countries along with
immuno-compromised individuals. “Traveler’s diarrhea” is caused by enterotoxigenic *E.
coli* (ETEC) where 2 enterotoxins are produced. Enterohemorrhagic *E. coli* (EHEC)
causes hemorrhagic colitis and serotype O157:H7 is the dominate serotype in this group and causes bloody diarrhea (Doyle, 1997).

**Description and history of E. coli O157:H7**

In 1982, *E. coli* O157:H7 (EHEC) was recognized as a human pathogen (Buchanan and Doyle, 1997; Byrd, 1999; Cheville, 1996; Feng, 1995). More than 100 strains have been identified with the toxin gene (Acheson, 1999). In 1987, the Shiga-like toxins of *E. coli* O157:H7 and *Shigella* were compared by Calderwood et al. (1987) and they found both bacteria shared the same receptor, structure, and mode of action. Some other disease causing serotypes include O26:H11, O13:H2, O11:H, and O113:H21 with a large outbreak occurring due to O11 in Australia (Acheson, 1999). More than 160 serotypes are known to cause disease in humans and over 200 in cattle. The bacterium is designated by its somatic O (polysaccharide) and flagellar H antigens (Buchanan and Doyle, 1997). According to Acheson (1999), *E. coli* O157:H7 obtained the Shiga-like toxin when it became infected with a bacteriophage that contained the Shiga toxin genes.

The majority of EHEC cases are caused by contaminated food and water, but person to person transmission has been reported (Benjamin and Datta, 1995). Because person to person transmission occurs, it is believed the infectious dose is low, less than or equal to 1,000 cells (Chen and Kasper, 1998; Labudde, 2002). This pathogen has been isolated from retail ground beef, pork, poultry, lamb and fecal samples from calves and heifers (Padhye and Doyle, 1991).
Pathogenicity

The animal and human gut is an ideal place for bacterial growth for some types of microorganisms and there can be as many as one trillion cells/g of digesta (Cornell University, 1998). *E. coli* O157:H7 adheres to epithelial cells in the lining of the intestinal tract and there are several stages to this adhesion. As the bacteria approach the intestinal cell surface, a loose association between the bacteria and cell is formed. Next, an “attaching and effacing lesion” occurs where the bacteria make and secrete a number of proteins that affect underlying human gut cells. Intimin is made by the bacteria and is expressed on the bacterial cell surface. This transmembrane bacterial protein mediates actin (part of the epithelial cell cytoskeleton) aggregation in cells attached to the organism by acting as hooks and creating intimin docking sites or receptors on the human host cells. Symptoms

More than 20,000 infections from *E. coli* occur each year (Cornell University, 1998). An estimated 2 to 7% of persons infected with *E. coli* O157:H7 will develop HUS and have renal failure (Besser et al., 1993). The incidence of infection by *E. coli* O157:H7 is 1 out of 10,000 people (Web MD, 1999). Symptoms of this illness start as short lived abdominal pains, cramping, and fever. After 1 to 2 d, non-bloody and bloody diarrhea develops and lasts 4 to 10 d with an average duration of 8 d (Tarr, 1995). Next hemolytic uremic syndrom (HUS) may develop 1 week after onset of diarrhea along with intravascular destruction of red blood cells (hemolysis), depressed platelet counts, lack of urine formation (anurina), swelling, and acute renal failure (Buchanan and Doyle, 1997).
Ten-percent of infected children less than 10 years of age suffer from HUS (Tarr, 1995). HUS is the leading cause of renal failure in U.S. children. In adults, thrombotic thrombocytopenic purpura (TTP) can develop along with neurological abnormalities. Symptoms and difficulties from TTP include seizures, coma, stroke, colonic perforation, pancreatitis and hypertension.

**Diagnosis**

This pathogen does not ferment sorbitol, hence the use of sorbitol MacConkey agar for distinction between it and generic *E. coli* by cultural methodology. Diagnosis of *E. coli* O157:H7 involves plating a stool specimen on sorbitol MacConkey agar that contains d-sorbitol instead of lactose as the carbon source. *E. coli* O157:H7, along with most other hemorrhagic *E. coli* strains, cannot utilize the d-sorbitol and colonies will remain colorless rather than the bright pink colonies characteristic of non-hemorrhagic strains (Feng, 1995). There is no known association between virulence of *E. coli* O157:H7 to its inability to ferment d-sorbitol (Padhye and Doyle, 1991; Tarr, 1995).

**Outbreaks**

Several hemorrhagic colitis outbreaks were recorded in the U.S. in 1982 and surveys done in the late eighties showed that 3.5% of ground beef, 1.5% of pork, 1.5% poultry and 2.0% lamb samples were contaminated with *E. coli* O157:H7, with hamburger being the most common source of infection. Since its recognition as a human pathogen in the 1980's, there have been more than a dozen outbreaks linked to *E. coli* O157:H7 and some of these are listed in Table 1.1 (Feng, 1995).
Prevention

In addition to consumers taking on more responsibility for prevention of foodborne diseases, several relatively cost effective, simple steps can be done by the food industry. Avoidance of fecal contamination as the meat is processed and adding a steam pasteurization cabinet or organic acid rinses are methods that have been used in attempts to minimize pathogens (Labudde, 2002). Other methods which will reduce the number of *E. coli* O157:H7 in the slaughter house are reducing the number of cattle shedding the bacteria and reducing the magnitude of the shedding. Animals fasting for longer than 24 h prior to slaughter is not an option because of the Humane Slaughter Act of 1958 (Labudde, 2002). There is a debate concerning what to feed pre-slaughter animals. Some suggest switching the diet from grain to hay 5 d prior to slaughter (Orr, 2003). This is considered advantageous to some since the hay does not provide residual starch to be fermented in the colon thus preventing exposure of bacteria to the acid conditions conducive for ATR which may increase survivability. However, others have shown that switching the diet yielded more fecal shedding than if the diet remained the same. Also, hay fed cattle shed *E. coli* O157:H7 longer than those fed grain (Orr, 2003).

**LISTERIA**

*Description and history of Listeria*

*Listeria* is a gram positive, nonfastidious, motile, non-sporeforming, non-acid fast, mesophilic, catalase positive, oxidase negative, urease negative pathogen seen in mammals (37 species), birds (17 species), fish and shellfish (U.S. Food And Drug
According to Leyer and Johnson (1993), 1-9% of the human population has *L. monocytogenes* in their feces. This pathogen targets the elderly, immuno-compromised individuals, pregnant women, fetuses and diabetics. *L. monocytogenes* caused the first recorded case of human listeriosis in 1985 although it was known for its illness in domestic animals since 1929 (Bremer et al., 1995). *L. monocytogenes* was the only known species until 1985 when others, such as *L. ivanovii* and *L. seeligeri*, were discovered. Three of the eight known strains cause human and animal infection.

*Listeria* is unique in that it is ubiquitous and able to not only survive freezing temperatures (-20°C), but also shows an increased virulence at low temperatures and is able to reproduce at 3°C even though its optimal temperature range is 30-37°C. These characteristics make *L. monocytogenes* a threat to ready-to-eat foods. It is also salt tolerant. Human to human transmission also occurs.

**Pathogenicity**

There are three steps in listeriosis infection. First the host cell is penetrated by *Listeria* followed by survival and multiplication of the pathogen. Finally, the target tissue is invaded by the bacterium. The principal site of human infection is the liver.

**Symptoms**

Listeriosis is characterized by septicemia, encephalitis, spontaneous abortions, stillbirths, flu-like symptoms, persistent fever, nausea, vomiting and diarrhea (U.S. Food
And Drug Administration, 1992a). Onset of disease is a few days to three weeks with gastrointestinal symptoms appearing within 12 h.

**Diagnosis**

Listeriosis can only be positively diagnosed by culturing the organism from blood, cerebrospinal fluid, or stool. Once the diagnosis is made, the treatment is usually penicillin or ampicillin. Gentamycin has been recommended by some experts, but its efficiency is questionable. Both septra and erythromycin have been shown to be effective in the treatment of *Listeria* (U.S. Food and Drug Administration, 1992a).

**Outbreaks**

The foods that can harbor *L. monocytogenes* include pasteurized milk, raw milk, soft cheese, coleslaw and pate (Centers for Disease Control and Prevention, 2002a). The majority of the cases are due to consumption of contaminated dairy and plant foods (Samelis and Metaxopoulos, 1999). Meat is also a culprit but less frequently. Table 1.2 highlights listeriosis outbreaks since 1980. The oral dose able to cause infection is unknown but believed to be low (fewer than 1,000 cells in susceptible individuals) (Samelis and Metaxopoulos, 1999; U.S. Food And Drug Administration, 1992a). Because of the seriousness of listeriosis and low infectious dose, many countries, including the U.S. have implemented a zero tolerance for *Listeria* in ready-to-eat foods. While this may be the case, food producers or processors are not required to test for *Listeria* except for ready-to-eat foods and being an ubiquitous organism, *Listeria* would probably be found in many of the samples albeit in low numbers.
**Prevention**

Prevention consists of thoroughly cooking raw food from animal sources, such as beef, pork or poultry (U.S. Food And Drug Administration, 1992a). Raw vegetables should be washed thoroughly before eating. Uncooked meats should be kept separate from vegetables and from cooked foods and ready-to-eat foods. Consumption of unpasteurized milk or foods made from unpasteurized milk should be avoided. Washing hands, knives, and cutting boards after handling uncooked foods should be practiced.

**SALMONELLA**

**Description and history of Salmonella**

*Salmonella* has been known to cause illness for over 100 years (Centers for Disease Control and Prevention, 2002b). An American scientist named Salmon discovered this bacterium and hence the name *Salmonella* (D’Aoust, 1997). More than 2,000 serotypes of human disease causing *Salmonella* have been identified (Doyle and Cliver, 1990) with serotypes Typhimurium and Enteritidis being among the most common ones encountered in the U.S. (Centers for Disease Control and Prevention, 2002b). This rod shaped, gram negative, non-spore-forming, facultative anaerobic, motile bacterium can be found in or on environmental sources that include water, soil, factory surfaces, kitchen surfaces, animal feces, raw meats, raw poultry, and raw seafood (Foster, 1993; U.S. Food And Drug Administration, 1992b).
Pathogenicity

*Salmonella* serotypes typically cause one of three diseases in humans: gastroenteritis (e.g., *S.* Typhimurium, *S.* Enteritidis, etc.), enteric fever (*S.* Typhi or *S.* Paratyphi) or an invasive systemic disease (*S.* Choleraesuis). The most common illness encountered in the U.S. is the gastroenteritis salmonellosis (duration 1-4 d) which is caused by 150 different serotypes (Doyle and Cliver, 1990). Less than 2.5% of human salmonellosis cases in the U.S. are due to *S.* typhi which causes typhoid fever. Enteric fever is also infrequent (<0.5%) in the U.S. Enteric and typhoid fever are caused by *S.* Paratyphi A, *S.* Schottmuelleri and *S.* Hirschfeldii and also share food vectors such as raw milk, raw salad, eggs and shellfish.

One source estimates that 40,000 cases of salmonellosis are annually reported in the U.S. and approximately 1,000 people die due to its effects (Centers for Disease Control and Prevention, 2002b). Another source estimates as many as 2-4 million cases per year occur (U.S. Food And Drug Administration, 1992b). The majority of the outbreaks are caused by consuming undercooked eggs, raw meat and poultry, unpasteurized milk, cross-contamination, and improper handling. Foods that have harbored *Salmonella* include frog legs, coconut, sauces and salad dressings, cake mixes, cream-filled desserts and toppings, dried gelatin, peanut butter, cocoa, and chocolate (U.S. Food and Drug Administration, 1992b). Most domestic pets including cats, dogs, reptiles, pigs and ducks can be carriers of *Salmonella*. Humans have also been carriers
for several days to months and this is especially critical when the carrier is a food handler employed in nursing homes and child-care centers.

To become infectious, the bacteria must pass from the gut lumen and penetrate into the epithelium of the small intestines where inflammation then occurs (U.S. Food and Drug Administration, 1992b). Evidence even suggests that an enterotoxin may be produced, but this has not yet been proven. Most people with the disease can rid themselves of the bacteria without any treatment. Onset of chronic related diseases like Reiter’s syndrome requires the bacterium to infect mucosal surfaces. While infectious dose is difficult to measure, several factors, such as type of food and strain, play a role. For a healthy individual the infectious dose is thought to be $10^8$ or $10^9$ cells (Cliver) but for immuno-compromised individuals the infectious dose may be as low as 1-10 cells (D’Aoust, 1997).

**Symptoms**

Salmonellosis results in vomiting, diarrhea, nausea, headache, fever and cramping within 12-72 h after consumption of contaminated food (Centers for Disease Control and Prevention, 2002b). Salmonellosis may develop in anyone but as with *Listeria*, children and elderly are more susceptible. Long term problems can develop from this infection. Postenteritis reactive arthritis and Reiter’s syndrome characterized by painful joints, irritation of the eyes and painful urination may appear after 3 weeks of infection and may persist for months to years after disease onset (U.S. Food and Drug Administration, 1992b). This can even lead to chronic arthritis (Centers for Disease Control and
Prevention, 2002b). Antibiotic treatment, such as ampicillin, gentamicin, trimethoprim/sulfamethoxazole or ciprofloxacin, is used when the disease has spread into the bloodstream. Some Salmonella, like Salmonella Typhimurium DT 104, have developed a resistance to antibiotics possibly due to the heavy use of antibiotics in animal feed (Centers for Disease Control and Prevention, 2002b).

**Diagnosis**

Diagnosis, made with a stool sample, requires several days for results. Once the presence of Salmonella is known, additional tests determine which serotype is involved and thus the proper antibiotic can be given, if necessary. Rapid methods are now available to food companies that give results for the presence of Salmonella within 48 h (U.S. Food and Drug Administration, 1992b).

**Outbreaks**

As seen in Table 1.3, many foods have been associated with salmonellosis. The majority of the outbreaks have been from eggs and plant foods (e.g., sprouts). The variety of food carriers makes testing and detection difficult.

**Prevention**

Preventative measures include: (1) washing hands well after using the restroom; (2) washing hands well after handling raw meat or poultry; (3) not eating raw or undercooked eggs, meat, or poultry; (4) not drinking unpasteurized milk; (5) disposing of dirty diapers properly; (6) washing hands well after changing diapers; (7) refrigerating
food promptly; (8) thawing foods properly and; (9) avoid cross contamination, especially among raw and cooked foods.

PHYSICAL PROPERTIES

Intrinsic factors are ones that are inherent to the food itself and examples of such factors include water activity, pH, oxidation-reduction potential, moisture content and nutrient content (Jay, 2000). Some of these factors will be defined and their roles in food spoilage and/or pathogen survival in food will be discussed.

Water Activity, \(a_w\)

Water activity is the ratio of vapor pressure of water in a material to the vapor pressure of pure water at the same temperature (Fontana, 2001). Perhaps the best definition is the “energy state of water in the food” and its “potential to act as a solvent and participate in chemical and physical reactions and growth of microorganisms” (Fontana, 2001). Pure water has an \(a_w\) of 1.00 since all of the water that is present is available for chemical reactions and microbial growth. As the water becomes bound in chemical bonds or is used by microorganisms, the vapor pressure of the solution decreases as does the calculated \(a_w\). The mathematical equation for \(a_w\) is \(p/p_o\) where \(p = \) vapor pressure of the solution (in a closed system) and \(p_o = \) vapor pressure of pure water (in a closed system). Controlling water activity in food for preservation purposes has been used for thousands of years despite the lack of scientific reasoning why it works. Sugar, salt, dehydration, and freezing are a few of the ways to alter the water activity of a food. Lowering the water activity is used to stabilize and protect the foods in regard to
microbiological reactions, chemical and physical properties, and rate of deteriorative reactions (Fontana, 2001). Reducing $a_w$ results in an increased lag phase in microbial cells which will in turn decrease the growth rate (Jay, 2000). Several factors control water activity and can be categorized as either osmotic or matric effects (Fontana, 2001). Colligative effects, capillary effects and surface interactions are such examples. Colligative effects include the dissolved salt or sugar and interaction with the water via dipole-dipole, ionic and hydrogen bonds. Capillary effects involve changes in the hydrogen bonding between water molecules. Surface interactions occur when water interacts directly with chemical groups on undissolved ingredients (starch and proteins) via dipole-dipole forces, ionic bonds ($\text{H}_3\text{O}^+$ or $\text{OH}^-$), van der Waals forces and hydrogen bonds (Fontana, 2001). Water activity, not water content, determines the lower limit of available water for microbial growth (Fontana, 2001). The lowest $a_w$ in which the majority of food spoilage bacteria can grow is 0.90 (Fontana, 2001). In general, gram negative bacteria require a higher $a_w$ than gram positive bacteria due to cell wall differences (Jay, 2000).

Water activity is temperature dependent and some products have an increased water activity with increasing temperature and vice versa. Foods with high water activities show negligible variation in water activity when compared at varying temperatures. Water activity can be measured using either chilled mirror dew point technology or relative humidity in combination with change in electrical resistance or
capacitance. Both methods vary in accuracy, repeatability, speed of measurement, stability in calibration and convenience of use (Fontana, 2001).

The importance of water activity related to the safety and shelf life of foods is evident by the fact that regulations by the U.S. Food and Drug Administration and U.S. Department of Agriculture references water activity in Good Manufacturing Practices and Hazardous Analysis and Critical Control Point plans. Studies have shown that at $a_w$ levels below 0.60 the available water is tightly bound so it is unavailable to most organisms.

In general, bacteria require a higher $a_w$ for growth compared to yeasts and molds. The $a_w$ for most fresh foods is 0.99 or greater. Water activity of dry foods is known to affect survival and thermal inactivation of *Salmonella*. Mattick et al. (2000) have shown that proliferation of *Salmonella* is inhibited at $a_w$ less than 0.93. Bearson et al. (1996) found that the $a_w$ of cereal would inhibit growth of *Salmonella*, but if the bacterium was already established on it, it would be able to survive.

**Moisture Content**

Moisture content is different from $a_w$ since it includes both bound and unbound water where as $a_w$ refers to unbound water only. There are several methods used to determine moisture content of a sample: forced oven draft, vacuum oven, microwave oven and infrared drying. Another method to determine the water content of a food product consists of weighing the food, drying it in a 105°C oven overnight, and then weighing the dried food. The AOAC Method 935.47 is yet another procedure to obtain
moisture content (AOAC, 1995). For oven-drying methodologies, the following equation is used to determine percent moisture on a wt./wt. basis.

\[
\% \text{ moisture} = \frac{(\text{wt. of wet sample} - \text{wt. of dry sample}) \times 100}{\text{wt. of wet sample}}
\]

*Color Measurements*

Color can be scientifically defined. In 1905, A.H. Munsell developed a system that measures color of an object in terms of hue, value, and chroma. While the system has been revised over time, it is still in use today. Others methods include the XYZ tristimulus values, Yxy color space, L*C*h, Hunter Lab color space, and L*a*b. The present CIE color space, also called L*a*b, is the most common color space for measuring an object based on the principle of color sensing by the human eye. This system was developed from the XYZ tristimulus values and Yxy color space systems. The “L” value reflects lightness and “a” and ”b” values are chromiticity (vividness) coordinates. A positive “a” value reflects red and a negative “a” value reflects green color, while a positive and negative b value reflects yellow and blue, respectively (DeMan, 1990).

*pH*

When discussing pH, words like acid, base, hydronium ion (H$_3$O$^+$) and hydroxide ion (OH$^-$) are often used. Acids are substances that increase hydronium ion concentration when added to water and bases are ones that increase hydroxide ion concentration (Ketchum, 1984). A scale was developed to measure pH which is defined as the negative logarithm of the hydrogen ion concentration (pH= - log [H$^+$]).
Undissociated acid acts as an antimicrobial agent (Adams and Hall, 1988). Lipophilic undissociated acid molecules penetrate the bacterium’s plasma membrane. In high pH cytoplasm, the acid dissociates to release protons and conjugate bases which in turn disrupt the membrane’s proton motive force disabling the energy yield and transport on which it depends. This is why pKa, and not solely pH play a role in the destructive nature of acids on bacteria. Acid resistance, acid habituation and acid tolerance response are three distinct conditions cells may undergo when exposed to acidic conditions.

**CELLULAR RESPONSES TO ACIDIC ENVIRONMENTAL STRESS**

In order to survive in the host, pathogenic bacteria must be able to overcome stresses such as the acidic stomach, physical barriers of epithelial cells that line the gastrointestinal tract and various immune defenses like the onslaught of macrophages. Beginning in the mouth, bacteria are exposed to digestive enzymes followed by exposure to low pH, volatile fatty acids, bile, and low oxygen in the small intestines (Gahan and Hill, 1999). Enterics thrive at a homeostasis of pH 7.6-7.8 and as long as the pH is within 1 unit in either direction, homeostasis is responsible for survival of the cell (Montville, 1997). pH homeostasis is dependent on how permeable the bacterial cell membrane is to the protons in the acid. Internal pumps either remove or introduce protons into the cell depending on whether they are exposed to acidic or alkaline conditions (Montville, 1997). Activated in acidic environments, the K-proton antiporters and Na-proton antiporters act as cellular pumps. Evidence supports that induction to acid tolerance depends on hydrogen ions crossing the outer membrane to activate a sensor in the periplasma or on
the periplasmic face of the cytoplasmic membrane with passage of protons being most probable via the pho E porin. Protons cross the outer membrane using this pho E system (Pinedo et al., 1987; Rowbury et al., 1992). In bacterial cells, the cytoplasmic membrane creates a barrier between external environment and cellular cytoplasm which regulates what enters and leaves the cell. These actions permit homeostasis of the cytoplasm. Whenever the cell’s homeostasis system cannot function to maintain a neutral pH, other systems within the cell are activated.

Acid resistance

Acid resistance (AR) response mechanism applies to cells in stationary phase that are grown in minimal media and acid challenged. Challenge pH values typical range from pH 2.0-2.5. There are three types of AR responses (Montville, 1997). The first response is activated by a brief exposure to glutamate prior to pH challenge. The second type of AR response requires extracellular glutamate during the challenge pH challenge. The alternate sigma factor controls this process. No induction is needed for this response, but several hours at low pH are required. For the third AR response, arginine is required during the pH challenge.

Acid Habituation

Acid habituation is often associated with log phase *E. coli* cells. If acid build up is gradual, then the cells may habituate. Buchanan and Edelson define acid habituation as exposure to moderate acidic conditions (pH 5.0) leading to withstanding more acidic pH values (<2.5) (Buchanan and Edelson, 1999). Habituation can occur rather rapidly. For
instance, at pH 5.0 organisms habituate within 7-10 min at 37°C (Rowbury et al., 1992; Rowbury and Hussain, 1987). Foster (2000) uses habituation to describe proliferation of the bacterium in nutrient broth that is exposed for a short period of time (7 min) to pH 3.

Acid Tolerance

Acid tolerance response (ATR) protects log phase cells during long term exposure to low pH. This reaction involves several steps. Media containing exponential phase cells is acidified to a moderate pH (near 5.5) for several hours and then exposed to a lethal pH (< 4.0). Two types of ATR have been described. Transiently induced ATR requires the Fe regulator, Fur. Once the proteins are activated, they remain unstable if not engaged at pH 3.3 for 20-30 min. The second type of ATR is referred to as sustained ATR which is dependent on rpoS. Growth in media with pH 7, followed by acidification at pH 4 achieves sustained ATR. Virulent bacterial strains exhibit sustained ATR and this process can occur rapidly, within 20 min. ATR can be explained in part by the cell’s ability to repair damaged DNA caused by high H⁺ concentration. ATR is growth phase dependent and requires the stress-specific sigma factor rpoS for full induction (Rowbury, 1995).

It is unclear what triggers induction of the rpoS but some believe it may be due to the slowing of cell division because of the encountered stress (Baik et al., 1996). Growth rate and phase of cells determine the expression of rpoS controlled regulon which aids in stressful situations (Jordan et al., 1999). There is both an exponential (log) and stationary phase ATR. Cells in stationary phase exhibit poor pH homeostasis but exhibit the highest
ATR (Jordan et al., 1999). In *E. coli* O157:H7, fifty acid shock proteins are induced in exponential phase cells (Baik et al., 1996). Of these proteins, 8 require the alternative sigma factor rpoS which is required for sustained ATR in *Salmonella*. Starved cells and stationary cells are more acid resistant than exponential cells. As with *E. coli* O157:H7, rpoS is responsible for ATR in *Salmonella*. The concentration of the acid, pH of the environment and dissociation constant of the chemical are factors that influence ATR. The unionized protonated form of the acid is more permeable to the cytoplasmic membrane than the ionized form (Baik et al., 1996). Exponential phase ATR occurs in both minimal and complex media. Cells are grown to mid-exponential phase and then the pH of the media is changed to pH 5. The “adapted” cells are then challenged at pH 2.5-3.5 for 1-4 h. This type of ATR is present in *E. coli*, *Listeria* and *Salmonella* but not in *Shigella flexneria*.

**Acid shock**

Acid shock is encountered when the cell goes straight from a neutral pH to one that is acidic (e.g.,<4.0). Cells undergoing ATR have phenotypical responses that aid in survival of bacterial cells exposed to extreme acidity (Brown et al., 1997). This coping method involves a two-stage process: adaptive period where the cells are exposed to a mild pH (5.0-6.0) followed by an acid challenge or shock exposure to pH below 4.0 (Garren et. al, 1997). Adapted cells resist acid damage to DNA better than unadapted (Rowbury, 1995).
Table 1.1. Selected pathogenic *E. coli* outbreaks, location of outbreak, source of outbreak, and number of confirmed cases per outbreak since 1982.

<table>
<thead>
<tr>
<th>Year</th>
<th>Place</th>
<th>Source of outbreak</th>
<th>Number of Cases</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>1982</td>
<td>Oregon</td>
<td>Hamburger at fast food</td>
<td>26</td>
<td>Doyle et al., 1997</td>
</tr>
<tr>
<td>1989-90</td>
<td>Montana</td>
<td>Drinking water</td>
<td>243</td>
<td>Doyle et al., 1997</td>
</tr>
<tr>
<td>1991</td>
<td>Oregon</td>
<td>Swimming water</td>
<td>21</td>
<td>Doyle et al., 1997</td>
</tr>
<tr>
<td>1991-92</td>
<td>Massachusetts</td>
<td>Unpasteurized apple cider-dropped apples</td>
<td>26</td>
<td>Keene et al., 1997</td>
</tr>
<tr>
<td>1993</td>
<td>Multi-state</td>
<td>Raw hamburger</td>
<td>731</td>
<td>Doyle et al., 1997</td>
</tr>
<tr>
<td>1994</td>
<td>Washington &amp; California</td>
<td>Pre-sliced dry fermented salami</td>
<td>23, 2 HUS</td>
<td>Tilden et al., 1996; Faith et al., 1998</td>
</tr>
<tr>
<td>1994</td>
<td>Virginia</td>
<td>Undercooked ground beef</td>
<td>20</td>
<td>Centers for Disease Control and Prevention, 1995a</td>
</tr>
<tr>
<td>1995</td>
<td>Oregon</td>
<td>Home-style jerky</td>
<td>5</td>
<td>Keene, et al., 1997</td>
</tr>
<tr>
<td>2000</td>
<td>Multi-state</td>
<td>Conagra</td>
<td>~ 30</td>
<td>Labudde, 2002</td>
</tr>
</tbody>
</table>
Table 1.2. Selected Listeria monocytogenes outbreaks, location of outbreak, source of outbreak, and number of confirmed cases since 1981.

<table>
<thead>
<tr>
<th>Year</th>
<th>Place</th>
<th>Source</th>
<th>Number of Cases</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>1981</td>
<td>Canada-1st North American</td>
<td>Coleslaw</td>
<td>41</td>
<td>Bahk and Marth, 1990</td>
</tr>
<tr>
<td>1983</td>
<td>Massachusetts</td>
<td>Pasteurized Milk</td>
<td>49</td>
<td>Bahk and Marth, 1990</td>
</tr>
<tr>
<td>1985</td>
<td>California</td>
<td>Mexican-style Cheese</td>
<td>100+</td>
<td>Bahk and Marth, 1990</td>
</tr>
<tr>
<td>1989-1990</td>
<td>United Kingdom</td>
<td>Pate</td>
<td>300</td>
<td>Rocourt and Cossart, 1997</td>
</tr>
<tr>
<td>2000</td>
<td>Multi-state</td>
<td>Deli Turkey</td>
<td>47</td>
<td>Centers for Disease Control and Prevention, 2002c</td>
</tr>
<tr>
<td>2002</td>
<td>United States</td>
<td>Turkey</td>
<td>46</td>
<td>Anonymous, 2003</td>
</tr>
</tbody>
</table>
Table 1.3. Selected *Salmonella* outbreaks, location place of outbreak, source of outbreak, and number of confirmed cases since 1974.

<table>
<thead>
<tr>
<th>Year</th>
<th>Place</th>
<th>Source</th>
<th>Number of Cases</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>1974</td>
<td>Navajo Reservation</td>
<td>Egg in potato salad</td>
<td>est 3,400</td>
<td>D’Aoust, 1997</td>
</tr>
<tr>
<td>1984</td>
<td>Canada</td>
<td>Cheese</td>
<td>est 2,700</td>
<td>D’Aoust, 1997</td>
</tr>
<tr>
<td>1985</td>
<td>Georgia</td>
<td>Turkey salad</td>
<td>100</td>
<td>Centers for Disease Control and Prevention, 1985</td>
</tr>
<tr>
<td>1991</td>
<td>Japan</td>
<td>Egg</td>
<td>10,000 +</td>
<td>D’Aoust, 1997</td>
</tr>
<tr>
<td>1995</td>
<td>Multi-state</td>
<td>Ice cream products</td>
<td>80</td>
<td>Centers for Disease Control and Prevention, 1994</td>
</tr>
<tr>
<td>1995</td>
<td>New Mexico</td>
<td>Beef jerky</td>
<td>93</td>
<td>Centers for Disease Control and Prevention, 1995a</td>
</tr>
<tr>
<td>1995</td>
<td>Nevada</td>
<td>Turkey &amp; dressing</td>
<td>7</td>
<td>Centers for Disease Control and Prevention, 1995b</td>
</tr>
<tr>
<td>1998</td>
<td>Midwest/East</td>
<td>Toasted oats</td>
<td>209</td>
<td>Centers for Disease Control and Prevention, 1998b</td>
</tr>
<tr>
<td>2002</td>
<td>Florida</td>
<td>Tomatoes</td>
<td>141</td>
<td>Centers for Disease Control and Prevention, 2002b</td>
</tr>
</tbody>
</table>
REFERENCES


CHAPTER 2

EFFECT OF VARIOUS PROCESSING PARAMETERS ON PATHOGEN REDUCTION OF HOME-STYLE BEEF JERKY

\[\text{\textsuperscript{1}}\]

\[\text{\textsuperscript{1}}\]Rose, R.A., J.A. Harrison, and M.A. Harrison. To be submitted to Journal of Food Protection.
ABSTRACT

Beef jerky is a popular dehydrated food. Due to foodborne illness cases linked to home prepared jerky during the 1980's and 1990's (4,5), questions arose concerning the safety of the product. There are numerous variations on how home-style beef jerky can be prepared. The objectives of this paper were to address several different safety concerns and aspects of home-style beef jerky. When applied to jerky strips, both sugar and salt marinades significantly increased the lethality of the drying process on inactivation of *Escherichia coli* O157:H7, *Listeria monocytogenes* and *Salmonella* spp. There was no significant difference in the reduction of *E. coli* O157:H7 populations inoculated on beef strips regardless of whether the cells were in direct or indirect contact with the liquid marinade during marination. The effectiveness of vertical air flow dehydration versus horizontal air flow dehydration on reducing the populations of *E. coli* O157:H7, *L. monocytogenes* and *Salmonella* spp. was determined along with the comparison of the final product’s physical properties. Among the physical properties measured, horizontal air flow reduced the water activity and moisture level of jerky more than vertical air flow. The water activity and percent moisture for strips dried with horizontal and vertical air flow were 0.679 and 0.735 and 20.04% and 23.06%, respectively. Lethality of moist versus dry heat applied as a post-dehydration step to inactivate *E. coli* O157:H7, *L. monocytogenes* and *Salmonella* spp. jerky strips was determined. Using dry heat is as effective as moist heat in the post-dehydration step. Factors that affect the outcome of the physical properties of jerky as well as the safety
aspects were revealed during this study and all of the steps that result in a safer jerky can be applied by a consumer with minimal effort for these scenarios.
INTRODUCTION

Dehydration, one of the oldest food preservation methods, reduces the water activity ($a_w$) of the food, which in turn reduces microbial growth and increases lag phase of bacteria. Synergistic antimicrobial effects can occur during beef jerky preparation due to the interaction between reduced product $a_w$, increased air temperature used for drying, and the variety of ingredients used in marination of jerky.

Several beef jerky studies have been done to help clarify the importance and significance of certain steps in the jerky process. Alternative, safer preparation methods were examined for home-style jerky preparation. However, producing safe jerky that also retains acceptable quality attributes is important. Lethality of *E. coli* O157:H7, *L. monocytogenes* and *Salmonella* as well as consumer acceptability and sensory attributes of jerky prepared by four methods (traditional, boil strips prior to drying, pre-cook strips to 71.1°C in an oven prior to drying, and heating strips to 71.1°C after drying) were examined. Of the four treatments, consumers preferred heating strips in the oven after drying even over the traditional method (10). The authors found that although the four treatments were significantly different in color, saltiness and texture, only texture appeared to influence overall consumer acceptability. Microbial challenge studies subjected *E. coli* O157:H7, *L. monocytogenes* and *Salmonella* spp. to the four treatments resulted in log population reductions of 5.8, 3.9 and 4.6, respectively. Oven treatment of strips after
drying reduced the pathogen populations by an additional 2 logs. A safer acceptable home-dried beef jerky can be produced by heating jerky strips to 71.1°C after drying.

A study by Albright et al. (1) investigated 4 different pre-treatments of whole strip jerky on \textit{E. coli}. After 10 h of drying, the treatment seasoned for 24 h at 4°C followed by submersion in pickle brine at 78°C for 90 s was shown to have the largest overall reduction and the highest pretreatment reduction (5.7-5.8 log CFU/cm²). The other pre-treatments consisting of: (1) boiling at 94°C for 15 s with marination at 4°C for 24 h; (2) immersing strips in a 1:1 vinegar and water solution for 20 s at 57.5°C with marination at 4°C for 24 h and; (3) marination at 4°C for 24 h and then immersing in a 1:1 vinegar and water solution for 20 s at 57.5°C, showed a 4.3-4.5, 4.9-5.2 and 4.7-4.8 log CFU/cm² reduction, respectively (1). This research would be more valuable if sensory testing of the various pre-treatments were conducted so that the application of the safest method could be applied by the consumer.

In a separate study using ground beef jerky that either contained or lacked a nitrite cure mix, \textit{E. coli} O157:H7, \textit{L. monocytogenes}, \textit{S. Typhimurium} populations were reduced by 2.5-4.0 logs CFU/g after 8 h of drying in samples that lacked the cure mix (11,12). When cure mix was added, the populations were reduced by at least 4 logs. Faith et al. (8) and Buege and Luchansky (3) have shown in previous jerky experiments that the fat content of the meat can influence the reduction of pathogens. Meat with higher percentages of fat provided extra protection for the pathogens that were present.
The interactions between all variables associated with home-style whole beef jerky lead to many questions related to safety and consumer friendly aspects that need to be investigated. When marinating, often only one side of the meat strip is submerged in the marinade. No previously reported studies determined the effect this has on safety of the finished product. Suggested recommendations have been made to incorporate a post-dehydration step using heat to further enhance pathogen reduction on jerky. For consumer acceptance, the physical properties of beef jerky made with safer recommendations should be similar to jerky made the more traditional way or similar to commercially available products. All of these concerns were addressed with the intention to apply what was learned to the consumer so a safer, quality jerky is produced.

MATERIALS AND METHODS

**Bacterial strains and inoculum preparation.** *E. coli* O157:H7 932 (clinical), E009 (beef), 204 P (pork), E0019 (cattle feces), and 380-94 (salami) and *L. monocytogenes* (Brie cheese isolate), Scott A (clinical isolate), LCDC #81-861 (coleslaw outbreak isolate), V7 (milk isolate) and 301 (cheddar cheese isolate) were obtained from the Center for Food Safety, The University of Georgia, Griffin, GA. *Salmonella* (*S. Typhimurium* 654, *S. Typhimurium* DT 104 H3380, *S. Typhimurium* DT 104 H3402, *S. California* and *S. Enteritidis*) were obtained from USDA/ARS, Athens, GA. All strains were preserved on Microbank™ beads (Pro-Lab Diagnostic, Austin, TX) frozen at -20°C. Each strain was activated in 9 ml portions of tryptic soy broth (TSB; Difco Labs, Division of Becton Dickinson and Co., Sparks, MD) at 37°C (statically) for 20-24 h. Each culture was
centrifuged for 20 min at 2,500 x g and the pellet re-suspended in 10 ml 0.1% peptone water (Bacto peptone, Difco Labs). The five strains of each bacterial type were pooled just prior to inoculation. One side of each beef strip was inoculated. One-hundred µl of each pool was inoculated on separate 1/3 portions of the strip surface to prevent the pathogen types from overlapping. Adhesion time was 30 min at room temperature under a laminar air flow hood unless noted otherwise. One ml of each pool was placed in 9 ml tubes containing 0.1% peptone and serially diluted to determine initial inoculum levels for each bacterial pool.

**Bacterial enumeration and enrichment.** For experiments requiring bacterial enumeration, sampling was done by placing a jerky strip in a sterile stomacher bag with 225 ml 0.1% peptone. It was then pummeled in a stomacher (TekMar model 400, Cincinnati, OH) for 2 min on high speed. Serial dilutions were made using 0.1% peptone buffer. Portions were spiral plated (Autoplate 4000; Exotech, Gaithersburg, MD) on bismuth sulfite agar (BSA; Difco Labs), *Listeria* selective agar (LSA; Oxoid; Basingstoke, Hampshire, England), and sorbitol MacConkey agar (SMAC; Oxoid) for *Salmonella*, *L. monocytogenes* and *E. coli O157:H7* enumeration, respectively. The plates were incubated for 24 h at 37°C before colony forming units were counted and then re-incubated and re-examined after an additional 24 h.

For the marination versus non-marination strips, subcultures of pummeled samples were enriched in the event that the populations of the pathogens were reduced to levels not detectable by direct plating. Enrichment consisted of inoculating 1 ml from the
stomacher bag into 9 ml portions of lactose broth (Difco Labs), UVM 

Listeria

enrichment broth base (Oxoid), and modified tryptic soy broth (modified TSB; Difco Labs) (10.0 g casamino acids, 1.5 g bile salts No.3, 6.0 g dibasic, anhydrous sodium phosphate and 1.35 g potassium phosphate per liter of TSB) for Salmonella, L. monocytogenes and E. coli O157:H7 enrichment, respectively. All three enrichment broths were incubated at 37°C for 18-24 h. After incubation, portions of the modified TSB cultures were streak plated onto SMAC plates. Plates were incubated at 37°C for 24 h and examined for the presence of representative colonies. Subcultures were also made from the lactose broth into selenite cystine (Difco Labs) and TT broth Hajna (Difco Labs) and from UVM Listeria enrichment broth into Fraser broth (Difco Labs) and then incubated at 37°C for 24 h. After incubation of the broths, portions were streak plated onto BSA, XLD (Difco Labs) and brilliant green agar (BGA; Difco Labs) for possible Salmonella spp. isolates and onto LSA for possible L. monocytogenes isolates. Plates were incubated at 37°C for 24 h and examined for the presence of representative colonies.

The identification of representative, presumptive isolates from the enrichment steps above were tested. Presumptive Salmonella spp. and E. coli O157:H7 isolates were identified using the Micro-ID™ identification system for Enterobacteriaceae (Remel, Lenexa, KS) as per manufacturer’s instructions. Salmonella isolates were serotyped with Salmonella O-antisera and E. coli isolates were serotyped with E. coli O157 and H7 antisera. (Difco Labs). Listeria isolates were identified using the Micro-ID™ Listeria system (Remel, Lenexa, KS) as per manufacturer’s instructions.
**Beef strip preparation.** Bottom round steak was purchased from a local Athens, GA supermarket. The edges of the beef were trimmed of visible fat and then sliced into 6.0 x 1.5 x 1.5 cm size strips.

**Marinade composition.** The marinated strips were stored for 16 h at 4°C in either a typical salt level marinade consisting of 60 ml (1/4 cup) soy sauce, 15 ml (1 Tbs.) Worcestershire sauce, 0.6 g (1/4 tsp.) black pepper, 1.25 g (1/4 tsp.) garlic powder, 4.35 g (1 tsp.) hickory smoked flavor salt and 1.5 g (½ tsp.) onion powder/ 454 g (1lb.) meat or a sugar based marinade containing 237 ml (1 cup) lite soy sauce, 5.86 g (1 tsp.) salt, 2.86 g (2 tsp.) hot pepper flakes, 2.66 g (1 tsp.) paprika, 9.04 g (2 tsp.) minced garlic and 44.4 ml (3 Tbs.) maple syrup/ 454 g (1 lb.) meat.

**Dehydrators.** Two home-style vertical air flow food dehydrators (model # 1000, American Harvest, Inc., Chaska, MN) were used to dehydrate both strip types. The dehydrators were preheated to 60°C prior to drying the strips. The internal air temperature and internal temperature of a strip for each dehydrator were monitored and recorded by a data recorder (model RD106 A, Omega, Stamford, CT) equipped with copper/constantan thermocouples (5TC-TT-T, Omega, Stamford, CT).
**Physical properties.** Water activity ($a_w$) was measured at $25 \pm 2^\circ C$ with a model CX2 Aqua Lab water activity meter (Decagon Devices, Inc., Pullman, WA). pH was measured by the surface method using a surface electrode and a Corning model 340 pH meter (Corning, Inc., Corning, NY). Percent moisture was determined by the AOAC Method 935.47 (AOAC International, 1995) and Quantab® titrator strips (0.5-1.0% Cl-) were used to measure percent sodium chloride (Environmental Test Systems, Inc., Elkhart, IN). However, modification to the instructions were required because the salt content in the jerky samples were higher than the strips could measure. As per the manufacturer’s instructions, a 10 g finely chopped sample was added to 90 ml of boiling water and stirred vigorously for 30 s, allowed to stand for 1 min and then stirred vigorously for another 30 s. The Quantab® reading was multiplied by 10 to calculate actual salt concentration in the sample. Maximum load and toughness (amount of energy necessary to break the sample) were assessed using a Warner-Bratzler shearing device until the total break of the sample. These force-deformation curves were measured by a texture analyzer model TA-XTZ (Texture Technologies Corp., Scarsdale, NY). The strips were placed inside the Warner-Bratzler shear blade and sheared perpendicular to the muscle. For each sample, two strips were sheared and each strip was sheared twice. The average force (N) of the four values was calculated. The area of a Minolta chroma meter (model CR-310) was used to measure $L^*a^*b$ values.
**Dehydrators.** The American Harvest, Inc. model # FD40 dehydrator (Chaska, MN) was used as a vertical air flow drier and the Equiflow model #7010 (B & J Industries, Marysville, WA) was used to obtain horizontal air flow.

**Marination versus non-marination.** Half of the beef strips used for this study were marinated with a salt based marinade, while the rest were not marinated. A vertical air flow dehydrator set at 60°C was used to dry the meat. Four different methods of preparation for marinated strips were examined: (1) dehydrated; (2) dehydrated and then placed in a 135°C oven (mechanical convection oven, model 18 EM; Precision Oven, Chicago, IL) for 10 min; (3) boiled for 5 min in marinade and dehydrated and; (4) placed in 135°C oven for 10 min and then dehydrated. Non-marinated strips were treated in the same fashion. For the non-marinated strips that were boiled, water was used in lieu of marinade. Strips were sampled after each step in the jerky making process. This was done in triplicate.

**Effect of direct versus indirect contact of marinade on E. coli O157:H7.** Four strips of meat were surface inoculated with the 5 strain pool of *E. coli* O157:H7 as described previously. Two of the strips were placed face down in a shallow pan containing the salt based marinade and two strips were placed face up in the pan. The pans were placed in a 4°C refrigerator for 8 h (+ 3 h) prior to enumeration of surviving *E. coli* O157:H7 populations on plate count agar (PCA, Difco Labs) and SMAC as described previously. This was done in triplicate.
Comparison of a sugar and salt marinade on physical properties of jerky prepared in a vertical and horizontal air flow dehydrator during various storage conditions.

Sugar and salt marinated jerky strips dried in either a 60°C horizontal or vertical air flow food dehydrator were prepared. Percent moisture, $a_w$ and pH were measured for raw beef, beef strips after marination and jerky after dehydration for both marination types. After dehydration, jerky strips were divided into the following groups for a storage study: (1) sugar marinated strips in ziplock® type bags stored at room temperature in a desiccator; (2) salt marinated strips stored in a ziplock® bag at room temperature in a desiccator; (3) sugar marinated strips stored in a Cryovac® bag placed inside a desiccator and stored at room temperature; (4) salt marinated strips stored in a Cryovac® bag placed inside a desiccator and stored at room temperature; (5) sugar marinated strips stored at 4.0°C in a Cryovac® bag; and (6) salt marinated strips stored at 4.0°C in a ziplock® bag. All samples were stored for 4 weeks prior to measuring $a_w$.

Effect of vertical vs. horizontal air flow on physical properties of beef jerky dried at various temperatures and comparison of various post-drying treatments. Beef strips inoculated with *E. coli* O157:H7 strains were dried in either a horizontal or vertical air flow food dehydrator with an internal temperature of either 54 or 60°C. Post-drying treatments resulted in an internal temperature of 71.1°C under moist or dry heat conditions. To examine the effect of dry versus moist heat for reduction of *E. coli* O157:H7 populations on beef jerky strips, the strips were post-inoculated in the same manner as described previously. The strips were then placed on a baking sheet. The
strips were exposed to one of three treatments. The control strips were placed on a baking sheet and placed directly into the oven. The second treatment had the strips and pan wrapped in aluminum foil. The last treatment consisted of the strips on the pan being wrapped in foil and 1 cup of water was placed in the foil with caution taken not to wet the strips. The strips were then placed in either a 93.3°C oven for 30 min. or a 135.0°C oven for 10 min. (mechanical convection oven, model 18 EM, Precision Oven, Chicago, IL). Samples were plated on PCA and SMAC at the following intervals; before marination, after marination, after dehydration, and after oven treatment. This experiment was done in triplicate.

**Physical properties of commercial brands of whole beef jerky.** Five commercially available beef jerky brands (A, B, C, D, E) were purchased from the Internet and a local Athens, GA grocery store to compare product $a_w$, pH, percent moisture, percent NaCl, and color (L*a*b). The brands were compared among themselves and with home-style whole strip beef jerky.

**Statistical analysis.** For the study comparing vertical and horizontal air flow, the average bacterial counts of three separate replications were transformed using the logarithmic transformation. Comparison was made using SAS which compared the log reduction of vertical versus horizontal air flow. The two dehydrator temperatures (54 or 60°C) were also compared among and between air flow types. The oven conditions (dry and moist) were also compared within and between air flow types. The physical properties measured for the five commercial brands of jerky were also analyzed with SAS (16). Analyses
were conducted using the PROC GLM procedure for SAS for both home-made and commercially available jerky.

RESULTS

Marination versus non-marination. There was a larger log population reduction for strips that were marinated compared to the non-marinated strips. The log bacterial population reduction for *E. coli*, *L. monocytogenes*, and *Salmonella* were 5.8, 4.1, and 4.6 for marinated strips and 4.0, 2.8, and 3.5 for the non-marinated strips, respectively (Tables 2.1, 2.2, 2.3). Marination in a salt-based marinade increased the lethal effect of drying for all three pathogens. For the strips that were boiled for 5 min. prior to drying, bacterial population of pathogens reached undetectable levels of *E. coli* O157:H7 and *L. monocytogenes* by direct plating (4.0 x 10^2) for both marinated and non-marinated strips (Tables 2.1 and 2.2). The bacterial log populations of *E. coli* and *L. monocytogenes* were reduced to undetectable levels (4.0 x 10^2) for both pre-heating in an oven at 163°C for 10 min and post-heating at 135°C in an oven for 10 min for marinated and non-marinated jerky strips (Tables 2.1 and 2.2). *Salmonella* populations were reduced on marinated strips that were treated in an oven to undetectable levels for both heat treated strips (Table 2.3).
Detectable *Salmonella* were still present for both pre-heating in an oven at 163°C for 10 min and post-heating at 135°C in an oven for 10 min for non-marinated strips (Table 2.3).

**Effect of direct versus indirect contact of marinade on E. coli O157:H7.** The average mesophilic aerobic populations for the strips that were placed in the marinade so the inoculated surface was not submerged in the marinade and plated on PCA was $1.87 \times 10^9$ CFU/strip. For those strips where the inoculated surface was in direct contact with the marinade, the populations were $1.62 \times 10^9$/strip. *E. coli* O157:H7 populations detected on the SMAC plates for strips that were not submerged and submerged in the marinade were $1.1 \times 10^7$ and $2.17 \times 10^6$, respectively. Strips exposed to direct contact with marinade resulted in *E. coli* O157:H7 CFU/strip populations that were ~ 1 log lower than strips with indirect contact. As would be expected, the non-selective PCA yielded higher populations than the SMAC plates.

**Comparison of a sugar and salt marinade on physical properties of jerky prepared in a vertical and horizontal air flow dehydrator during various storage conditions.**

Sugar marinated jerky had an $a_w$ of 0.65 and a pH of 5.27. Salt marinated jerky had an $a_w$ of 0.75 and a pH of 5.64. The drying time for the sugar marinated jerky to reach the same visual endpoint was approximately 3 h longer than that for the salt marinated jerky.

For the comparison of air flow direction and various storage conditions, there was no noticeable difference in the $a_w$ of sugar or salt marinated samples stored under various conditions. The various storage conditions for 4 weeks and their respective water activities are presented in Table 2.4.
Effect of vertical vs. horizontal air flow on physical properties of whole beef jerky dried at various temperatures and comparison of various post-drying treatments.

Dehydrator temperature (54 or 60°C) had a significant effect (p<0.05) on pH and percent moisture of the final product. Airflow direction significantly (p<0.05) affected water activity of the final product, with strips dried at 54.4°C using horizontal air flow producing a product with a lower water activity (0.679) as compared to the vertical flow (0.735). For strips dried at 60°C, the water activity of strips dried with horizontal and vertical air flow were 0.665 and 0.706, respectively. Airflow type also significantly affected moisture levels (p<0.05) of the product, with strips dried at 54.4°C using horizontal air flow resulting in a lower moisture level (20.04%) versus vertical flow (23.06%) (Table 2.6). This same trend was observed in strips dried at 60°C with horizontally dried strips yielding a 16.03% moisture and vertical yielding 20.24% moisture. Strips dried at 60°C had less available water than those dried at 54°C (Table 2.7).

Comparison of E. coli O157:H7 population reduction and physical properties for dry and moist heat on post-dehydrated jerky strips. Results for the comparison of moist versus dry heat on reduction of E. coli O157:H7 showed that the average log population reduction determined on SMAC plates for jerky strips exposed to dry or moist heat after dehydration was 5.73 for both treatments (Table 2.5). Tables 2.6, 2.7, and 2.8 summarize the physical properties for the various air flows, dehydrator temperatures, oven temperatures, and heating types. Tables 2.9 and 2.10 contain the texture analysis.
results for the air flow and various post treatments. For strips dried at 54.4°C using vertical air and heated in a 93.3°C oven, more force (kg) was used to sheer the samples compared with the strips that were placed in the 135°C oven. For strips dried at 54.4°C, time required to sheer strips for both air flow directions, despite post oven temperature, were within 8.53 and 11.43 s. The area (kg.s) under the force-deformation curve was not noticeably different for all strips dried at 54.4°C and 60.0°C with values ranging from 46.10-62.84 kg.s and 39.01-59.65 for strips dried at 54.4°C and 60.0°C, respectively.

**Comparison of commercial brands of whole beef jerky.** Table 2.8 shows the values of the physical properties for both the commercial and home-style jerky. For all sampled jerky, there was no significant difference seen for the physical property (p>0.05).

**DISCUSSION**

There are numerous variations on preparation methods used for home-style beef jerky. A number of factors involved, if done marginally, raise food safety concerns. In addition there have been several jerky outbreaks since the early 1980's (4,5). This paper addresses several different safety aspects of home-style jerky. The effects of marination on the survival of *E. coli* O157:H7, *L. monocytogenes* and *Salmonella* spp. on whole beef jerky strips were determined. The salt in the marinade aids in reducing *a_w* of the product, which in turn aids in maintaining safety and shelf life of jerky (15).

Soy sauce, fermented soybeans or wheat, consists of 5% ethanol, sodium benzoate, and lactic acid or acetic acid and has a pH of 4.5 (14). It is often the main ingredient in many jerky marinades and the antimicrobial effects of the lactic or acetic
acid help reduce pathogen populations on whole jerky strips. During the home-style marination process, often the meat is not fully submerged in the marinade. To determine if direct contact with the marinade is required for pathogen inhibition or population reduction, a study was done that compared *E. coli* O157:H7 populations on strips that were not submerged and submerged in the marinade during marination. Results from non-submerged and submerged strips were not noticeably different. The findings from this experiment support the current recommendations such as those found in food preservation guides such as *So Easy to Preserve* (2) for jerky marination in which the marinade may not cover the entire strip. The use of more marinade to entirely cover the strip does not enhance the effort to reduce *E. coli* populations on the product.

In a comparison of effect a salt-based marinate or a sugar-based marinate had on physical attributes of the beef jerky, the salt marinated product had a higher $a_w$ than the sugar marinated product. The sugar marinated beef strips required a longer drying time (3 h longer) compared to the salt marinated strips. A recipe calling for a sugar based marinade may need more drying time to achieve the same level of safety as the salt marinated strips. Jerky is a product with a long shelf life assuming the storage conditions do not introduce moisture into the dehydrated meat. A storage study was conducted involving both sugar and salt marinated jerky where the final products were stored for 4 weeks under various conditions. The different combinations of storage temperatures, bag types and storage conditions examined in this study showed there was no difference in the water activity ($a_w$) over time related to the type of marinade used to prepare the product.
An experiment was designed to compare the physical properties of jerky prepared in either a vertical or horizontal dehydrator dried at either 54 or 60°C. Horizontal air flow resulted in a lower water activity and lower percent moisture level than vertical flow. As the temperature increases, the humidity decreases and the osmotic pressure increases (7,9). Dehydration at the higher temperature resulted in lower water activity.

Strips were post-contaminated with *E. coli* O157:H7 and effect of dry versus moist heat on the pathogen inactivation was recorded. Some studies have shown that moist heat is more detrimental than dry heat due to the faster rate of protein denaturation that occurs with moist heat (13). Dry heat is less lethal and relies on dehydration and oxidation to kill the cells. Dry heat also requires higher temperatures and a longer heating time compared to wet heat (9). For this product and preparation process, data does not support the notion that moist heat has a greater lethal effect on *E. coli* O157:H7 when heated in an oven.

A comparison of the physical properties of commercially available whole strip beef jerky was made within the brands and with home-style whole strip beef jerky. Statistical analysis showed no significant difference (p>0.05) for *a*, percent moisture, percent sodium chloride, pH and color measurements (L*a*b).

Many factors contribute to the safety of home-style beef jerky. While one factor alone may not be significant for safety, the synergistic effects of the various factors contribute to the overall safety of the product. As new methods of jerky marination, preparation, dehydration and storage are introduced to consumers, the acceptability by the
consumer is an important aspect. Both the consumer acceptability and safety must be considered when evaluating current and alternative methods for home-style jerky.
ACKNOWLEDGMENTS

This project was supported financially in part from a grant from CSREES and USDA and by the Georgia Agricultural Experiment Stations.
REFERENCES


formed beef jerky prepared at levels of 5 and 20% fat and dried at 52, 57, 63, or 68°C in a home-style dehydrator. *Inter. J. Food Microbiol.* 41:213-221.


Table 2.1. Populations of *E. coli* O157:H7 (log CFU/strip) on marinated and non-marinated beef jerky strips for four different preparation treatments.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Marinated</th>
<th>Non-marinated</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Pre-treatment</td>
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</tr>
<tr>
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<tr>
<td>4&lt;sup&gt;f&lt;/sup&gt;</td>
<td>6.39</td>
<td>5.89</td>
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</table>

<sup>a</sup> Strips dehydrated at 60°C.

<sup>b</sup> This step of the jerky processing was not performed for this treatment.

<sup>c</sup> Populations were below detectable level (4.0 x 10²).

<sup>d</sup> Strips boiled for 5 min prior to drying at 60°C.

<sup>e</sup> Strips pre-heated in a 163°C oven for 10 min prior to drying at 60°C.

<sup>f</sup> Strips dehydrated at 60°C followed by heating in a 135°C oven for 10 min.
Table 2.2. Populations of *L. monocytogenes* (log CFU/strip) on marinated and non-marinated beef jerky strips for four different preparation treatments.

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<sup>a</sup> Strips dehydrated at 60°C.
<sup>b</sup> This step of the jerky processing was not performed for this treatment.
<sup>c</sup> Populations were below detectable level (4.0 x 10<sup>2</sup>).
<sup>d</sup> Strips boiled for 5 min prior to drying at 60°C.
<sup>e</sup> Strips pre-heated in a 163°C oven for 10 min prior to drying at 60°C.
<sup>f</sup> Strips dehydrated at 60°C followed by heating in a 135°C oven for 10 min.
Table 2.3. Populations of *Salmonella* (log CFU/strip) on marinated and non-marinated beef jerky strips for four different preparation treatments.

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<tr>
<td>Marinated</td>
<td>6.24</td>
<td>na&lt;sup&gt;b&lt;/sup&gt;</td>
<td>na&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.76</td>
</tr>
<tr>
<td>Non-marinated</td>
<td>6.24</td>
<td>&lt; 0.60&lt;sup&gt;d&lt;/sup&gt;</td>
<td>na&lt;sup&gt;b&lt;/sup&gt;</td>
<td>&lt; 0.60&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>6.24</td>
<td>na&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.28</td>
<td>1.85</td>
</tr>
<tr>
<td></td>
<td>6.24</td>
<td>na&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.84</td>
<td>2.05</td>
</tr>
</tbody>
</table>

<sup>a</sup> Strips dehydrated at 60°C.
<sup>b</sup> This step of the jerky processing was not performed for this treatment.
<sup>c</sup> Strips boiled for 5 min prior to drying at 60°C.
<sup>d</sup> Populations were below detectable level (4.0 x 10<sup>2</sup>).
<sup>e</sup> Strips pre-heated in a 163°C oven for 10 min prior to drying at 60°C.
<sup>f</sup> Strips dehydrated at 60°C followed by heating in a 135°C oven for 10 min.
Table 2.4. Average $a_w$ after 4 weeks of storage of whole strip beef jerky made with either a sugar or salt-based marinade.

<table>
<thead>
<tr>
<th>Storage Treatments</th>
<th>Sugar Marinated strips</th>
<th>Salt Marinated strips</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ziplock®/ Room temperature/ Desiccator</td>
<td>0.656</td>
<td>0.634</td>
</tr>
<tr>
<td>Cryovac®/ Room temperature/ Desiccator</td>
<td>0.682</td>
<td>0.570</td>
</tr>
<tr>
<td>Cryovac®/ Room temperature</td>
<td>0.656</td>
<td>0.670</td>
</tr>
<tr>
<td>Ziplock®/ 4°C</td>
<td>0.621</td>
<td>0.594</td>
</tr>
<tr>
<td>Ziplock®/ Room temperature</td>
<td>ns&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.638</td>
</tr>
</tbody>
</table>

<sup>a</sup>This storage treatment was not done for sugar marinated strips.
Table 2.5. Average log populations and log reduction for *E. coli* O157:H7 on beef jerky strips that were post-treated with dry or moist heat applied.

<table>
<thead>
<tr>
<th>Sample time</th>
<th>Log population</th>
<th>Log population reduction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before marinade</td>
<td>7.80</td>
<td>-</td>
</tr>
<tr>
<td>After marinade&lt;sup&gt;a&lt;/sup&gt;</td>
<td>7.47</td>
<td>0.33</td>
</tr>
<tr>
<td>After dehydration&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.74</td>
<td>5.06</td>
</tr>
<tr>
<td>After dehydration and oven heating&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.01</td>
<td>5.79</td>
</tr>
<tr>
<td>After dehydration and oven heating using dry heat&lt;sup&gt;d&lt;/sup&gt;</td>
<td>2.08</td>
<td>5.72</td>
</tr>
<tr>
<td>After dehydration and oven heating using moist heat&lt;sup&gt;e&lt;/sup&gt;</td>
<td>2.08</td>
<td>5.72</td>
</tr>
</tbody>
</table>

<sup>a</sup> After marination for 16 (+/−2 h) at 4°C.
<sup>b</sup> Strips dehydrated at 60°C.
<sup>c</sup> Dehydrated strips heated in a 163°C oven for 10 min.
<sup>d</sup> Dehydrated strips wrapped in aluminum foil and heated in a 163°C oven for 10 min.
<sup>e</sup> Dehydrated strips wrapped in aluminum foil with water added to the package and heated in a 163°C oven for 10 min.
Table 2.6. Comparison of air flow direction, post-dry oven temperatures and heating conditions on properties of whole beef strips dehydrated at 54.4 °C.

<table>
<thead>
<tr>
<th>Air flow direction</th>
<th>Post-dry oven temperature (°C)</th>
<th>Post-dry heating conditions</th>
<th>$a_w$</th>
<th>pH</th>
<th>Moisture (%)</th>
<th>NaCl (%)</th>
<th>L</th>
<th>Color$^a$</th>
<th>$a$</th>
<th>$b$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vertical</td>
<td>na$^b$</td>
<td>na$^b$</td>
<td>0.735</td>
<td>5.47</td>
<td>23.06</td>
<td>5.70</td>
<td>25.11</td>
<td>3.43</td>
<td>5.26</td>
<td></td>
</tr>
<tr>
<td>Vertical</td>
<td>93.3</td>
<td>Dry</td>
<td>0.696</td>
<td>5.65</td>
<td>21.31</td>
<td>6.30</td>
<td>25.22</td>
<td>4.01</td>
<td>5.50</td>
<td></td>
</tr>
<tr>
<td>Vertical</td>
<td>93.3</td>
<td>Moist</td>
<td>0.715</td>
<td>5.53</td>
<td>21.96</td>
<td>5.73</td>
<td>24.99</td>
<td>3.79</td>
<td>5.45</td>
<td></td>
</tr>
<tr>
<td>Vertical</td>
<td>135.0</td>
<td>Dry</td>
<td>0.714</td>
<td>5.57</td>
<td>22.50</td>
<td>5.70</td>
<td>25.84</td>
<td>3.90</td>
<td>6.13</td>
<td></td>
</tr>
<tr>
<td>Vertical</td>
<td>135.0</td>
<td>Moist</td>
<td>0.736</td>
<td>5.78</td>
<td>22.93</td>
<td>5.17</td>
<td>23.96</td>
<td>3.60</td>
<td>4.98</td>
<td></td>
</tr>
<tr>
<td>Horizontal</td>
<td>na$^b$</td>
<td>na$^b$</td>
<td>0.679</td>
<td>5.55</td>
<td>20.04</td>
<td>6.36</td>
<td>26.07</td>
<td>4.52</td>
<td>6.42</td>
<td></td>
</tr>
<tr>
<td>Horizontal</td>
<td>93.3</td>
<td>Dry</td>
<td>0.657</td>
<td>5.48</td>
<td>19.59</td>
<td>7.92</td>
<td>26.72</td>
<td>4.05</td>
<td>6.28</td>
<td></td>
</tr>
<tr>
<td>Horizontal</td>
<td>93.3</td>
<td>Moist</td>
<td>0.681</td>
<td>5.58</td>
<td>21.07</td>
<td>5.37</td>
<td>25.15</td>
<td>3.54</td>
<td>5.03</td>
<td></td>
</tr>
<tr>
<td>Horizontal</td>
<td>135.0</td>
<td>Dry</td>
<td>0.661</td>
<td>5.65</td>
<td>18.66</td>
<td>8.88</td>
<td>25.84</td>
<td>4.26</td>
<td>5.94</td>
<td></td>
</tr>
<tr>
<td>Horizontal</td>
<td>135.0</td>
<td>Moist</td>
<td>0.687</td>
<td>5.60</td>
<td>21.24</td>
<td>4.73</td>
<td>24.92</td>
<td>3.81</td>
<td>5.47</td>
<td></td>
</tr>
</tbody>
</table>

$^a$ Color measurements include lightness (L) values of 0 to 100, redness (a) values of -50 to +50 and yellowness (b) values of -50 to +50.

$^b$ This treatment was not post-treated.
Table 2.7. Comparison of air flow direction, post-oven temperatures and heating conditions on properties of whole beef strips dehydrated at 60.0°C.

<table>
<thead>
<tr>
<th>Air flow direction</th>
<th>Post-dry oven temperature (°C)</th>
<th>Post-dry heating conditions</th>
<th>$a_w$</th>
<th>pH</th>
<th>Moisture (%)</th>
<th>NaCl (%)</th>
<th>L</th>
<th>Color$^a$</th>
<th>a</th>
<th>b</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vertical</td>
<td>na$^b$</td>
<td>na$^b$</td>
<td>0.706</td>
<td>5.68</td>
<td>20.24</td>
<td>6.33</td>
<td>25.10</td>
<td>3.86</td>
<td>5.58</td>
<td></td>
</tr>
<tr>
<td>Vertical</td>
<td>93.3</td>
<td>Dry</td>
<td>0.717</td>
<td>5.70</td>
<td>20.96</td>
<td>5.49</td>
<td>25.50</td>
<td>3.83</td>
<td>5.74</td>
<td></td>
</tr>
<tr>
<td>Vertical</td>
<td>93.3</td>
<td>Moist</td>
<td>0.714</td>
<td>5.92</td>
<td>20.99</td>
<td>5.96</td>
<td>25.11</td>
<td>3.64</td>
<td>6.66</td>
<td></td>
</tr>
<tr>
<td>Vertical</td>
<td>135.0</td>
<td>Dry</td>
<td>0.695</td>
<td>5.74</td>
<td>21.39</td>
<td>4.77</td>
<td>25.51</td>
<td>3.63</td>
<td>5.61</td>
<td></td>
</tr>
<tr>
<td>Vertical</td>
<td>135.0</td>
<td>Moist</td>
<td>0.691</td>
<td>5.89</td>
<td>21.93</td>
<td>5.34</td>
<td>28.27</td>
<td>3.21</td>
<td>6.05</td>
<td></td>
</tr>
<tr>
<td>Horizontal</td>
<td>na$^b$</td>
<td>na$^b$</td>
<td>0.665</td>
<td>5.58</td>
<td>16.03</td>
<td>7.45</td>
<td>24.96</td>
<td>4.14</td>
<td>5.66</td>
<td></td>
</tr>
<tr>
<td>Horizontal</td>
<td>93.3</td>
<td>Dry</td>
<td>0.661</td>
<td>5.62</td>
<td>17.65</td>
<td>7.60</td>
<td>23.72</td>
<td>3.77</td>
<td>4.80</td>
<td></td>
</tr>
<tr>
<td>Horizontal</td>
<td>93.3</td>
<td>Moist</td>
<td>0.681</td>
<td>5.75</td>
<td>18.22</td>
<td>5.74</td>
<td>24.60</td>
<td>3.40</td>
<td>5.14</td>
<td></td>
</tr>
<tr>
<td>Horizontal</td>
<td>135.0</td>
<td>Dry</td>
<td>0.668</td>
<td>5.68</td>
<td>17.58</td>
<td>7.82</td>
<td>25.03</td>
<td>4.24</td>
<td>5.79</td>
<td></td>
</tr>
<tr>
<td>Horizontal</td>
<td>135.0</td>
<td>Moist</td>
<td>0.670</td>
<td>5.62</td>
<td>17.53</td>
<td>5.62</td>
<td>25.34</td>
<td>3.49</td>
<td>4.87</td>
<td></td>
</tr>
</tbody>
</table>

$^a$ Color measurements include lightness (L) values of 0 to 100, redness (a) values of -50 to +50 and yellowness (b) values of -50 to +50.

$^b$ This treatment was not post-treated.
Table 2.8. Comparison of air flow direction, post-oven temperatures and heating conditions on properties of whole beef strips dehydrated at 60.0°C and commercially available beef jerky.

<table>
<thead>
<tr>
<th>Post-dry oven temperature (°C)</th>
<th>Post-dry heating conditions</th>
<th>$a_w$</th>
<th>pH</th>
<th>Moisture (%)</th>
<th>NaCl (%)</th>
<th>$L$</th>
<th>Colora</th>
<th>a</th>
<th>b</th>
</tr>
</thead>
<tbody>
<tr>
<td>Home-style, vertical</td>
<td>na</td>
<td>0.706</td>
<td>5.68</td>
<td>20.24</td>
<td>6.33</td>
<td>25.10</td>
<td>3.86</td>
<td>5.58</td>
<td></td>
</tr>
<tr>
<td></td>
<td>93.3 Dry</td>
<td>0.717</td>
<td>5.70</td>
<td>20.96</td>
<td>5.49</td>
<td>25.50</td>
<td>3.83</td>
<td>5.74</td>
<td></td>
</tr>
<tr>
<td></td>
<td>93.3 Moist</td>
<td>0.714</td>
<td>5.92</td>
<td>20.99</td>
<td>5.96</td>
<td>25.11</td>
<td>3.64</td>
<td>6.66</td>
<td></td>
</tr>
<tr>
<td></td>
<td>135.0 Dry</td>
<td>0.695</td>
<td>5.74</td>
<td>21.39</td>
<td>4.77</td>
<td>25.51</td>
<td>3.63</td>
<td>5.61</td>
<td></td>
</tr>
<tr>
<td></td>
<td>135.0 Moist</td>
<td>0.691</td>
<td>5.89</td>
<td>21.93</td>
<td>5.34</td>
<td>28.27</td>
<td>3.21</td>
<td>6.05</td>
<td></td>
</tr>
<tr>
<td>Home-style, horizontal</td>
<td>na</td>
<td>0.665</td>
<td>5.58</td>
<td>16.03</td>
<td>7.45</td>
<td>24.96</td>
<td>4.14</td>
<td>5.66</td>
<td></td>
</tr>
<tr>
<td></td>
<td>93.3 Dry</td>
<td>0.661</td>
<td>5.62</td>
<td>17.65</td>
<td>7.60</td>
<td>23.72</td>
<td>3.77</td>
<td>4.80</td>
<td></td>
</tr>
<tr>
<td></td>
<td>93.3 Moist</td>
<td>0.681</td>
<td>5.75</td>
<td>18.22</td>
<td>5.74</td>
<td>24.60</td>
<td>3.40</td>
<td>5.14</td>
<td></td>
</tr>
<tr>
<td></td>
<td>135.0 Dry</td>
<td>0.668</td>
<td>5.68</td>
<td>17.58</td>
<td>7.82</td>
<td>25.03</td>
<td>4.24</td>
<td>5.79</td>
<td></td>
</tr>
<tr>
<td></td>
<td>135.0 Moist</td>
<td>0.670</td>
<td>5.62</td>
<td>17.53</td>
<td>5.62</td>
<td>25.34</td>
<td>3.49</td>
<td>4.87</td>
<td></td>
</tr>
<tr>
<td>Commercial brand A</td>
<td>na</td>
<td>0.790</td>
<td>5.88</td>
<td>29.49</td>
<td>5.28</td>
<td>26.67</td>
<td>13.68</td>
<td>4.78</td>
<td></td>
</tr>
<tr>
<td>Commercial brand B</td>
<td>na</td>
<td>0.753</td>
<td>5.54</td>
<td>28.58</td>
<td>6.57</td>
<td>23.99</td>
<td>6.95</td>
<td>2.72</td>
<td></td>
</tr>
<tr>
<td>Commercial brand C</td>
<td>na</td>
<td>0.787</td>
<td>5.67</td>
<td>29.27</td>
<td>6.07</td>
<td>27.30</td>
<td>11.59</td>
<td>4.23</td>
<td></td>
</tr>
<tr>
<td>Post-dry oven temperature (°C)</td>
<td>Post-dry heating conditions</td>
<td>$a_w$</td>
<td>pH</td>
<td>Moisture (%)</td>
<td>NaCl (%)</td>
<td>L</td>
<td>Color$^a$</td>
<td>a</td>
<td>b</td>
</tr>
<tr>
<td>-------------------------------</td>
<td>----------------------------</td>
<td>-------</td>
<td>----</td>
<td>--------------</td>
<td>---------</td>
<td>------</td>
<td>-----------</td>
<td>---</td>
<td>----</td>
</tr>
<tr>
<td>Commercial brand D</td>
<td>na</td>
<td>0.629</td>
<td>5.62</td>
<td>16.44</td>
<td>5.85</td>
<td>29.63</td>
<td>20.57</td>
<td>6.64</td>
<td></td>
</tr>
<tr>
<td>Commercial brand E</td>
<td>na</td>
<td>0.584</td>
<td>5.60</td>
<td>16.47</td>
<td>7.32</td>
<td>27.35</td>
<td>5.26</td>
<td>8.15</td>
<td></td>
</tr>
</tbody>
</table>

$^a$ Color measurements include lightness (L) values of 0 to 100, redness (a) values of -50 to +50 and yellowness (b) values of -50 to +50.

$^b$ For commercially bought jerky, these parameters do not apply. For home-style jerky, this treatment was not post-treated.
Table 2.9. Textural analysis using the Warner-Bratzler method for jerky strips dehydrated at 54.4°C with either vertical or horizontal air and post treated with dry heat or moist heat at either 93.3 or 135°C.

<table>
<thead>
<tr>
<th>No post treatment</th>
<th>Vertical</th>
<th></th>
<th></th>
<th>Horizontal</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Dry</td>
<td>Moist</td>
<td></td>
<td>Dry</td>
<td>Moist</td>
<td></td>
</tr>
<tr>
<td></td>
<td>93.3°C</td>
<td>135°C</td>
<td>93.3°C</td>
<td>93.3°C</td>
<td>135°C</td>
<td>93.3°C</td>
</tr>
<tr>
<td>Force (kg)</td>
<td>19.33±2</td>
<td>20.62 ± 1.68</td>
<td>20.26 ± 5.08</td>
<td>20.66 ± 3.10</td>
<td>19.43 ± 2.77</td>
<td>25.90 ± 3.21</td>
</tr>
<tr>
<td></td>
<td>± 2.60</td>
<td>± 1.3 ± 2.13</td>
<td>± 5.08 ± 4.48</td>
<td>± 3.10 ± 1.30</td>
<td>± 2.77 ± 3.21</td>
<td>± 5.21 ± 1.64</td>
</tr>
<tr>
<td>Time (s)</td>
<td>8.39 ± 0.02</td>
<td>11.43 ± 4.35</td>
<td>8.70 ± 0.25</td>
<td>8.69 ± 0.02</td>
<td>8.89 ± 0.26</td>
<td>8.56 ± 0.41</td>
</tr>
<tr>
<td></td>
<td>± 0.02</td>
<td>± 0.19 ± 0.25</td>
<td>± 0.12 ± 0.41</td>
<td>± 0.02 ± 0.25</td>
<td>± 0.26 ± 0.41</td>
<td>± 0.25 ± 0.43</td>
</tr>
<tr>
<td>Area (kg.s)</td>
<td>46.10 ± 5.76</td>
<td>53.05 ± 23.10</td>
<td>50.80 ± 16.75</td>
<td>56.45 ± 16.96</td>
<td>54.76 ± 2.78</td>
<td>62.84 ± 15.72</td>
</tr>
<tr>
<td></td>
<td>± 0.02</td>
<td>± 12.67 ± 16.75</td>
<td>± 21.57 ± 21.57</td>
<td>± 2.78 ± 16.96</td>
<td>± 15.72 ± 22.13</td>
<td>± 14.32 ± 22.13</td>
</tr>
</tbody>
</table>

* Means ± standard deviation.
Table 2.10. Textural analysis using the Warner-Bratzler method for jerky strips dehydrated at 60.0°C with either vertical or horizontal air and post treated with dry heat or moist heat at either 93.3 or 135°C.

<table>
<thead>
<tr>
<th></th>
<th>Vertical</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No post</td>
<td>Dry</td>
<td>Moist</td>
<td>No post</td>
</tr>
<tr>
<td></td>
<td>treatment</td>
<td>93.3°C</td>
<td>135°C</td>
<td>93.3°C</td>
</tr>
<tr>
<td>Force (kg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>21.64±2</td>
<td>20.25</td>
<td>18.48</td>
<td>23.69</td>
</tr>
<tr>
<td></td>
<td>± 2.75</td>
<td>± 5.68</td>
<td>± 3.08</td>
<td>± 3.94</td>
</tr>
<tr>
<td>Time (s)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>± 1.58</td>
<td>± 1.76</td>
<td>± 2.02</td>
<td>± 2.10</td>
</tr>
<tr>
<td>Area (kg.s)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>45.93</td>
<td>53.55</td>
<td>39.01</td>
<td>54.21</td>
</tr>
</tbody>
</table>

* Means ± standard deviation.
CHAPTER 3

FATE OF *ESCHERICHIA COLI* O157:H7, *LISTERIA MONOCYTOGENES* AND

*SALMONELLA* SPP. IN REDUCED SODIUM BEEF JERKY

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1Rose, R.A., J.A. Harrison, and M.A. Harrison. To be submitted to *Journal of Food Protection*. 

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ABSTRACT

Interest in low-sodium food products necessitates re-examination of home preservation processes relying in part on salt for antimicrobial effects. The fates of *Escherichia coli* O157:H7, *Listeria monocytogenes*, and *Salmonella* spp. in reduced sodium home-style beef jerky were determined. Ground or whole beef strips, with different salt levels, were inoculated with the pathogens. Samples were either dried in a 60°C dehydrator or heated to an internal temperature of 71.1°C prior to drying in a 60°C dehydrator. Populations were determined at time 0 and 2 h intervals until dry. Reductions of pathogens were greater in ground beef jerky with non-reduced salt levels compared to that with reduced salt levels, and in most cases, greater reduction (1.0-1.5 logs) was observed for ground beef strips heated prior to drying. However, at time intervals during the drying stage for whole and ground beef jerky, there were differences in the number of viable microorganisms when salt levels were compared, with less lethality in the reduced salt samples. Ground beef strips made with reduced sodium resulted in less pathogen reduction. Therefore, preparing ground beef jerky with reduced salt is a potential food safety hazard since the lower salt level did not reduce the pathogen population as much as the regular salt marinade. For dried whole jerky strips, there generally were no significant differences (p>0.05) in pathogen populations between the non-reduced and reduced salt marinade in the end product. The results from this study support the importance of the antimicrobial effect of sodium chloride in particular products on the pathogens used in this experiment.
INTRODUCTION

The use of salt (sodium chloride) in the preservation of meat products has been a widely accepted preservation method throughout recorded history. The antimicrobial properties of salt are well established, and it is a widely used ingredient in the preparation of beef jerky. The Surgeon General’s Dietary Guidelines for Americans (9) recommends a moderate intake of sodium. An abundance of low- or no-sodium products in the marketplace attracts consumers who are searching for products and new methods of preparation to help them reduce their intake of sodium. The desire to make low- or no-sodium products at home is an outgrowth of this movement. Home processes that were once considered to be safe due to antimicrobial effects of salt may need to be examined due to the growing popularity of low- and no-sodium recipes for home processed foods.

Outbreaks of foodborne illness due to Salmonella in beef jerky (3) and E. coli O157:H7 in venison jerky (7), coupled with the fact that a variety of preparation methods and drying procedures abound, raises concern over the safety of processed meat products made in the home. Discrepancies in temperatures and amounts of ingredients with preservative effects indicate the need for investigation. The inactivation of selected pathogens in beef jerky prepared with salt concentrations that were typically used have been evaluated previously (5, 6).
The objective of this investigation was to determine the fate of *E. coli* O157:H7, *L. monocytogenes*, and *Salmonella* spp. on home-style ground and whole beef jerky strips prepared with non-reduced and reduced salt levels.

**MATERIALS AND METHODS**

The investigation examined the effect of two drying methods (drying uncooked samples and drying pre-cooked samples that had reached an internal temperature of 71.1°C prior to drying) and two salt levels (non-reduced and reduced salt) on three different organisms (*E. coli* O157:H7, *L. monocytogenes*, and *Salmonella* spp.) for each type of meat (whole and ground beef strips). The trials for the ground beef strips and whole beef jerky were conducted separately with three replications on three different days for each type of meat. The meat for the three replications came from the same batch of meat for both product types.

**Bacterial strains and inoculum preparation.** *L. monocytogenes* Brie (cheese isolate), Scott A (clinical isolate), LCDC #81-861 (coleslaw outbreak isolate), V7 (milk isolate) and 301 (cheddar cheese isolate) and *E. coli* O157:H7 932 (clinical), E009 (beef), 204 P (pork), E0019 (cattle feces), and 380-94 (salami) were obtained from the Center for Food Safety, The University of Georgia, Griffin, GA. *Salmonella* (S. Typhimurium 654, S. Typhimurium DT 104 H3380, S. Typhimurium DT 104 H3402, S. California and S. Enteritidis) were obtained from USDA/ARS, Athens, GA. All strains were preserved on Microbank™ beads (Pro-Lab Diagnostics, Austin, TX) frozen at -20°C. Each strain was activated in 9 ml portions of tryptic soy broth (TSB; Difco Labs, Division of Becton
Dickinson and Co., Sparks, MD) at 37°C for 20-24 h, followed by transferring 1 ml of
culture to 120 ml TSB that were incubated at 32°C for 18-24 h. To wash each culture,
they were centrifuged for 20 min at 2,500 x g, spent TSB decanted, the pellet was re-
suspended in 20 ml Butterfield’s phosphate buffer (0.1M, pH 7.0) and then re-
centrifuged. After decanting the buffer, cultures were suspended in separate sterile tubes
containing 10 ml Butterfield’s phosphate buffer and then serially diluted to approximately
10^8 CFU/ml. The five strains of each bacterial type were pooled just prior to inoculation.
One ml of each pool was placed in 9 ml tubes containing 0.1% peptone and serially
diluted to determine initial inoculum levels for each bacterial pool.

**Whole strip beef jerky preparation and analysis.** Beef, loin cut, was obtained from
the Department of Food Science and Technology’s food processing laboratory and stored
at -20°C. Prior to inoculation, the beef was placed in a 4°C incubator for approximately
12 h to slightly thaw. The edges of the beef were trimmed of visible fat and then sliced to
approximately 15.0 x 1.5 x 1.5 cm strips. One side of each strip was inoculated with 100
µl of each pool on 1/3 portions of the strip to prevent the pathogen types from
overlapping. Samples were incubated at 4°C overnight (16 h ± 2 h) to allow for microbial
adhesion to the beef. The marinade used to represent one with regular levels of sodium
consisted of 118.5 ml soy sauce, 29.6 ml Worcestershire sauce, 9.8 g garlic salt, 9.8 g
black pepper and 9.8 ml liquid smoke/1,820 g of meat. The reduced sodium marinade
contained 118.5 ml low-salt soy sauce, 29.6 ml 65% less sodium Worcestershire sauce,
9.8 g garlic powder in lieu of garlic salt, 9.8 g black pepper and 9.8 ml liquid smoke/1,820 g of meat.

When sampled, each strip was placed in a sterile stomacher bag containing 100 ml of Butterfield’s phosphate buffer and stomached (TekMar model 400, Cincinnati, OH) for 30 s on regular speed. Portions were spread plated on bismuth sulfite agar (BSA; Difco Labs), *Listeria* selective agar (LSA) (Oxoid; Basingstoke, Hampshire, England), and sorbitol MacConkey agar (SMAC, Oxoid) for *Salmonella, Listeria* and *E. coli* O157:H7, respectively and incubated at 32°C for 24 h before counting colony forming units. The plates were then re-incubated and were re-examined after an additional 24 h incubation.

**Ground beef jerky preparation and analysis.** Lean ground beef (90% lean, pH 5.83) obtained from the Department of Food Science and Technology’s food processing laboratory was frozen immediately upon grinding and stored at -20°C. Prior to inoculation, meat was placed in a 4°C incubator for approximately 18 h to thaw. The spices and seasonings were added to 1,820 g (4 lb.) ground beef at the following levels: 19.6 g seasoning salt, 9.8 g salt, 4.9 g garlic salt, 2.5 g red pepper, 9.8 ml liquid smoke, 19.6 ml Worcestershire and 19.6 ml soy sauce. The reduced sodium mixture contained 19.6 g salt-free seasoning, 4.9 g garlic powder, 2.5 g red pepper, 9.8 ml liquid smoke, 19.6 ml Worcestershire sauce (having 65% less sodium than regular Worcestershire sauce) and 19.6 ml low-salt soy sauce. The regular salt mixture contained 6,490 mg of sodium while the reduced salt mixture contained 207 mg of sodium.
The ground beef was inoculated with 1 ml of each bacterial strain and mixed for 1 min using a mixer set on high speed (model K5-A, Hobart Corporation, Troy, OH). Jerky strips (33.5 x 2.5 x 0.75 cm) were made using The Beef Jerky Works™ (BJW#1P, American Harvest, Inc., Chaska MN). To determine initial microbial load, 25 g portions of beef were weighed, placed in a sterile stomacher bag with 100 ml Butterfield’s phosphate buffer and stomached (TekMar model 400, Cincinnati, OH) for 30 s on normal speed. Serial dilutions of the samples were prepared using Butterfield’s phosphate buffer. Portions were spread plated on BSA, LSA, and SMAC for *Salmonella*, *Listeria* and *E. coli* O157:H7, respectively and incubated at 32°C for 24 h before counting colony forming units. They were re-examined after an additional 24 h of incubation.

Subcultures of all pummeled samples in this study were enriched in the event that the populations of the three pathogens were reduced to undetectable levels (8.0 x 10^1) by direct plating. One ml portions of each sample were inoculated into 9 ml portions of TSB (Difco Labs). After incubation at 32°C for 24 h, the appropriate selective media (as listed previously) were streaked in order to detect presumptive viable *Salmonella*, *E. coli* O157:H7 or *L. monocytogenes*. The plates were incubated at 32°C for 24 h before observing for typical colony growth for the three pathogens used in this study.

The identification of representative, presumptive isolates from the enrichment steps above were tested. Presumptive *Salmonella* spp. and *E. coli* O157:H7 isolates were identified using the Micro-ID™ identification system for Enterobacteriaceae (Remel,
Lenexa, KS) as per manufacturer’s instructions. *Salmonella* isolates were serotyped with *Salmonella* O-antisera and *E. coli* isolates were serotyped with *E. coli* O157 and H7 antisera. (Difco Labs). *Listeria* isolates were identified using the Micro-ID™ *Listeria* system (Remel, Lenexa, KS) as per manufacturer’s instructions.

**Dehydration and heat treatments.** Inoculated samples were either placed directly into a 60°C pre-heated horizontal air flow food dehydrator (model #7010; B&J Industries, Marysville, WA) or heated in a convection oven (GCA mechanical convection oven, model 18 EM; Precision Oven, Chicago, IL) pre-heated to 93°C until the internal temperature of the strips reached 71.1°C. Internal temperature of the samples was monitored continuously during cooking with thermocouples and a Speedomax strip chart recorder (Type T-68, Leeds & Northup, North Wales, PA). After cooking, strips were placed in the pre-heated dehydrator and sampled at 0, 2, 4, and 6 h of drying at 60°C. Sampling intervals for the uncooked strips placed directly in the dehydrator were at 0, 2, 4, 6 and 8 h of drying. Two strips were analyzed at each sample time with the bacterial counts being averaged over both strips.

**Percent moisture and sodium chloride analysis.** Moisture content of jerky was determined by the AOAC Method 935.47 (1). Quantab® titrator strips (0.5-1.0% Cl-) were used to measure percent sodium chloride of the product (Environmental Test Systems, Inc., Elkhart, IN).

**Statistical analysis.** The experimental design was a complete block for each type of meat consisting of a factorial arrangement of the different combinations of drying
methods, salt levels and organisms. The sources of variation included the effects for drying method, salt level, organism and the interaction terms. The bacterial counts were transformed using the logarithmic transformation. Regression analysis was utilized to evaluate the rate of reduction in log bacterial counts over time. Due to the fact that the sampling periods for the uncooked and pre-cooked strips were not directly comparable, analysis of variances were conducted separately for the initial and final sampling periods. All analyses were conducted using the PROC GLM procedure in SAS (8).

RESULTS

Meat types dried under similar conditions had similar moisture content in the finished product (Tables 3.1 and 3.2). In general, salt concentration was higher in whole beef jerky than in ground beef jerky, both initially and throughout the drying period. This difference is partially due to the fact that the recipes for the two jerky types are not identical. As salt concentration increased, moisture content decreased (Tables 3.1 and 3.2).

The average log population for \( E. coli \), \( L. monocytogenes \), and \( Salmonella \) on cooked and uncooked ground beef jerky strips made with non-reduced and reduced salt levels are in Tables 3.3, 3.4, and 3.5, respectively. The average log population for \( E. coli \), \( L. monocytogenes \), and \( Salmonella \) on cooked and uncooked whole beef jerky strips made with low and regular salt levels are in Tables 3.3, 3.4, and 3.5, respectively. The analysis of variance for each meat type indicated no significant differences (p>0.05) among the main effects or interactions for the initial starting conditions.
The preliminary analysis of variance results of the bacterial population for the final sampling times for the ground beef strips indicated highly significant main effects for drying method (p=0.0001), organism (p=0.0057), and salt level (p=0.0017) with all the interactions being non-significant (p>0.05). The results for the whole beef strips indicated a highly significant effect for drying method (p=0.0001). In regressing the rate of reduction in log bacterial populations, it was found to be predominantly significantly linear (p<0.05) trends over time for each salt level within each drying method and type of meat. The R² values ranged from 0.29 to 0.77.

The primary focus of this research investigation was to determine the effect of salt level on bacterial survival at the final sampling time. Due to the fact that it was expected to have highly significant differences between drying methods on bacterial survival and that the residual mean square errors between drying types within the meat types were not homogenous, separate analyses were conducted for each drying method within each meat type. The observed significance levels for differences between the salt levels for each drying method within meat type were: uncooked ground beef strips p=0.0514, pre-cooked ground beef strips p=0.015, uncooked whole beef strips p=0.133 and pre-cooked whole strips p=0.810. No significant differences between salt levels for the whole beef strips were detected. The differences among the types of organisms revealed significant differences only for the uncooked ground beef strips.
DISCUSSION

_E. coli_ O157:H7 was more easily inactivated than _L. monocytogenes_ and _Salmonella_ spp. All organisms were more easily inactivated on whole strips where contamination is usually confined to the surface unlike the ground strips with contamination spread throughout. Pre-cooking whole strips prior to drying resulted in a decrease in _E. coli_ O157:H7 populations of 0.5-1.5 logs and _Salmonella_ populations decreased by 2 logs more than uncooked strips. The log reductions were similar for _Listeria_ regardless of whole strips being cooked or not. Regardless of the salt levels, there was no difference in population reductions of _Listeria_ and _Salmonella_ in whole beef jerky strips. _E. coli_ populations were slightly lower in whole strips that contained non-reduced salt levels. From this study, it is apparent that reducing the salt level for the ground beef samples resulted in less log bacterial population reduction. Uncooked samples had higher log bacterial populations compared to pre-cooked strips, especially for the ground beef strips. Uncooked ground beef strips made with regular salt levels had final log populations of 2.5, 4.86, and 3.09 for _E. coli_, _L. monocytogenes_, and _Salmonella_, respectively. The uncooked strips containing reduced salt levels had final log populations of 4.10, 4.96, and 4.44 for _E. coli_, _L. monocytogenes_, and _Salmonella_, respectively. Due to the lack of pathogen inactivation, it would be unadvisable for preparation of ground beef jerky unless the strips were pre-cooked, regardless of salt level. Salt contributes to the lethality of the drying process for ground beef jerky.
Hurdle technology is often used as a combined effort to control unwanted microorganisms. However, not all studies have shown that combinations of antimicrobial methods have a positive result. A study done by Casey and Condon (2) showed how the combination of sodium chloride and acid pH was less effective than acid pH alone on population reductions of *E. coli* O157:H45. The presence of sodium chloride reduced the bactericidal effect of lactic acid on exponential cells when grown in pH 4.2 media with 4% sodium chloride (simulating the salt concentration in fermented sausage) resulting in a $10^3$ fold higher survival rate when sodium chloride and acidic pH were used together. The sodium chloride is believed to counteract the acidification by organic acids (2).

Previously, Clavero and Beuchat (4) conducted a similar experiment but their conclusions did not support the concept of sodium chloride providing a protective effect on the cells exposed to low acid. This discrepancy in results may be due to the fact that Clavero and Beuchat used higher percent sodium chloride and incubation occurred at 5, 20, 30, and 37°C whereas Casey and Condon used 4% sodium chloride and incubated at 37°C only.

The safety of jerky relies on many factors. Salt plays a vital role in reducing water activity and lowering percent moisture which aids in the safety of the final product. In some cases the salt has shown to enhance survival of certain pathogens. The reduction of sodium in jerky marinades may contribute to pathogen survival so this practice should be avoided.
ACKNOWLEDGMENTS

This project was supported financially in part by a grant, validation of jerky processing and small-scale and home processors, funded by CSREES and USDA and by the Georgia Agricultural Experiment Stations.
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60:1139-1141.

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Table 3.1. Percent moisture and percent sodium chloride content of regular and reduced salt ground beef jerky during preparation by either drying at 60°C or by preheating jerky to 71.1°C before drying at 60°C.

<table>
<thead>
<tr>
<th>Time (h)</th>
<th>Dehydrated strips</th>
<th>Pre-cooked and dehydrated strips</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Regular</td>
<td>Reduced</td>
</tr>
<tr>
<td></td>
<td>% Moisture</td>
<td>% NaCl</td>
</tr>
<tr>
<td>Pre-cook</td>
<td>ns*</td>
<td>ns</td>
</tr>
<tr>
<td>0</td>
<td>68.99</td>
<td>1.12</td>
</tr>
<tr>
<td>2</td>
<td>55.28</td>
<td>ns</td>
</tr>
<tr>
<td>4</td>
<td>38.78</td>
<td>ns</td>
</tr>
<tr>
<td>6</td>
<td>32.46</td>
<td>ns</td>
</tr>
<tr>
<td>8</td>
<td>22.78</td>
<td>3.89</td>
</tr>
</tbody>
</table>

* ns: not sampled
Table 3.2. Percent moisture and percent sodium chloride content of regular and reduced salt whole beef jerky during preparation by either drying at 60°C or by preheating jerky to 71.1°C before drying at 60°C.

<table>
<thead>
<tr>
<th>Time</th>
<th>Dehydrated strips</th>
<th></th>
<th>Pre-cooked and dehydrated strips</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Regular</td>
<td>Reduced</td>
<td>Regular</td>
<td>Reduced</td>
</tr>
<tr>
<td></td>
<td>% Moisture</td>
<td>% NaCl</td>
<td>% Moisture</td>
<td>% NaCl</td>
</tr>
<tr>
<td>Pre-cook</td>
<td>ns*</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
</tr>
<tr>
<td>0</td>
<td>70.74</td>
<td>2.67</td>
<td>70.15</td>
<td>1.10</td>
</tr>
<tr>
<td>2</td>
<td>42.65</td>
<td>ns</td>
<td>42.68</td>
<td>ns</td>
</tr>
<tr>
<td>4</td>
<td>20.45</td>
<td>ns</td>
<td>25.36</td>
<td>ns</td>
</tr>
<tr>
<td>6</td>
<td>17.20</td>
<td>ns</td>
<td>14.99</td>
<td>ns</td>
</tr>
<tr>
<td>8</td>
<td>13.10</td>
<td>6.35</td>
<td>14.59</td>
<td>3.88</td>
</tr>
</tbody>
</table>

* ns: not sampled.
Table 3.3. The average log population of *E. coli* O157:H7 for the initial and final sampling time for ground and whole beef strips during preparation by either drying at 60°C or by preheating jerky to 71.1°C before drying at 60°C.

<table>
<thead>
<tr>
<th>Population</th>
<th>Ground Beef Strips</th>
<th>Whole Beef Strips</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Dehydrate Only</td>
<td>Pre-cook &amp; Dehydrate</td>
</tr>
<tr>
<td></td>
<td>Regular  Reduced</td>
<td>Regular  Reduced</td>
</tr>
<tr>
<td>Initial</td>
<td>8.12  7.87</td>
<td>7.35  7.42</td>
</tr>
<tr>
<td></td>
<td>(5.36)a</td>
<td>(6.80)a</td>
</tr>
<tr>
<td>Final</td>
<td>2.50  4.10</td>
<td>1.40  2.07</td>
</tr>
</tbody>
</table>

*a* Values in parentheses are the population log count immediately after preheating strips to 71.1°C for 10 min.

*b* Indicates below detection level
Table 3.4. The average log population of *L. monocytogenes* for the initial and final sampling time for ground and whole beef strips during preparation by either drying at 60°C or by preheating jerky to 71.1°C before drying at 60°C.

<table>
<thead>
<tr>
<th></th>
<th>Ground Beef Strips</th>
<th>Whole Beef Strips</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Dehydrate Only</td>
<td>Pre-cook &amp; Dehydrate</td>
</tr>
<tr>
<td>Population</td>
<td>Regular</td>
<td>Reduced</td>
</tr>
<tr>
<td>Initial</td>
<td>7.20</td>
<td>7.10</td>
</tr>
<tr>
<td></td>
<td>(5.17)a</td>
<td>(6.42)a</td>
</tr>
<tr>
<td>Final</td>
<td>4.86</td>
<td>4.96</td>
</tr>
</tbody>
</table>

*a* Values in parentheses are the population log count immediately after preheating strips to 71.1°C for 10 min.

*b* Indicates below detection level
Table 3.5. The average log population of *Salmonella* for the initial and final sampling time for ground and whole beef strips during preparation by either drying at 60°C or by preheating jerky to 71.1°C before drying at 60°C.

<table>
<thead>
<tr>
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<th>Ground Beef Strips</th>
<th></th>
<th>Whole Beef Strips</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Dehydrate Only</td>
<td>Pre-cook &amp; Dehydrate</td>
<td>Dehydrate Only</td>
<td>Pre-cook &amp; Dehydrate</td>
</tr>
<tr>
<td>Population</td>
<td>Regular</td>
<td>Reduced</td>
<td>Regular</td>
<td>Reduced</td>
</tr>
<tr>
<td>Initial</td>
<td>7.23</td>
<td>7.65</td>
<td>7.00</td>
<td>7.50</td>
</tr>
<tr>
<td></td>
<td>(3.78)(^a)</td>
<td>(6.33)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Final</td>
<td>3.77</td>
<td>3.09</td>
<td>1.40</td>
<td>2.77</td>
</tr>
</tbody>
</table>

\(^a\) Values in parentheses are the population log count immediately after preheating strips to 71.1°C for 10 min.
CHAPTER 4

SURVIVAL OF ACID-ADAPTED AND NONADAPTED *ESCHERICHIA COLI*,

*LISTERIA MONOCYTOGENES* AND *SALMONELLA* SPP. ON GROUND OR

WHOLE BEEF JERKY

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1Rose, R.A., J.A. Harrison, and M.A. Harrison. To be submitted to *Journal of Food Protection*.
ABSTRACT

The objective of this paper was to monitor and compare the survival of acid-adapted and nonadapted *Escherichia coli* O157:H7, *Listeria monocytogenes*, and *Salmonella* spp. on ground and whole beef jerky strips during the home-style jerky process. Each organism and meat type was compared separately and analyzed using a split-plot type experimental design. Samples were taken at time 0, 2, 4, 6, and 10 h for ground beef strips. Maximum log reduction was first observed after 6 h for all three pathogens in ground beef strips. For ground beef strips, the maximum mean log reduction over the entire 10 h drying process was higher for acid-adapted *E. coli* (6.22 logs) and *Salmonella* (4.73) compared to nonadapted *E. coli* (5.30 logs) and *Salmonella* (3.96). However, only the treatments for *E. coli* populations were significantly (p<0.05) different. In ground beef strips, the maximum log reduction for *L. monocytogenes* was greater in the nonadaptive strains (4.51) as compared to acid-adapted (4.28) strains after 10 h of drying. Sampling of whole beef strips was done at the following intervals: after inoculation, after marination (T:0), 4, 8, 12, and 14 h. Maximum log reduction was first observed after 8 h for whole strips. For whole beef strips, the log reduction was almost identical for strips inoculated with *Salmonella*. Log reduction for acid-adapted and nonadapted *E. coli* O157:H7 were 5.25 and 5.13, respectively. For *L. monocytogenes*, log populations declined by 4.81 logs for acid-adapted and 4.87 logs for nonadapted cells. The results from this study show there is no significant (p>0.05) difference in survival of *L. monocytogenes* and *Salmonella* acid-adapted and nonadapted cells on ground or whole beef home-style jerky. However, there was a significant difference for ground beef strips.
inoculated with *E. coli* O157:H7 with acid-adapted populations having a greater reduction after 10 h of drying.
INTRODUCTION

Exposure of foodborne bacteria to mild or severe acidic conditions frequently occurs as a result of the acids that are either found in or artificially added to foods. Whenever the cell’s pH homeostasis system cannot function to maintain a neutral pH, other systems within the cell are activated (9). These alternate systems are very complex and are growth dependent. Each system functions uniquely. Acid resistance (AR), acid habituation (AH) and acid tolerance response (ATR) are systems that allow survival by some bacteria under acidic conditions (1, 2, 3, 7, 13, 14). A recent concern that has become a food safety issue is the possibility that pathogenic cells can become tolerant or adapted to acid which may enhance survival upon exposure to acidic foods and defenses of the human body (e.g., stomach acidity and digestive enzymes). Acid adaptation in *E. coli* O157:H7, *L. monocytogenes* and *Salmonella* has been demonstrated in some laboratory studies (1, 2, 3, 7, 8, 11, 13, 14) and in food systems such as beef jerky and juices (4, 5, 6, 14).

Beef jerky is a popular dehydrated food. During home-style preparation, the meat is often marinated in an acidic marinade which may allow any bacteria that are present on the meat to become adapted to the acidic environment. This adaptation may enhance survival of pathogenic bacteria, especially on a food product like jerky that typically does not have a cooking step to inactivate unwanted bacteria. The objective of this paper was to monitor and compare the survival of acid-adapted and nonadapted *E. coli* O157:H7, *L. monocytogenes* and *Salmonella* spp. on ground and whole beef jerky strips during the home-style jerky process.
MATERIALS AND METHODS

Bacterial strains and inoculum preparation. E. coli O157:H7 932 (clinical), E009 (beef), 204 P (pork), E0019 (cattle feces), and 380-94 (salami) and L. monocytogenes (Brie, Scott A, LCDC, V7, 301) were obtained from the Center for Food Safety, The University of Georgia, Griffin, GA. Salmonella (S. Typhimurium 654, S. Typhimurium DT 104 H3380, S. Typhimurium DT 104 H3402, S. California and S. Enteritidis) were obtained from USDA/ARS, Athens, GA. All strains were preserved on Microbank™ beads (Pro-Lab Diagnostics, Austin, TX) frozen at -20°C. Each strain was activated in 9 ml portions of tryptic soy broth (TSB; Difco Labs, Division of Becton Dickinson and Co., Sparks, MD) at 37°C for 20-24 h. The method of Buchanan and Edelson (2) was used to obtain acid-adapted and nonadapted bacterial populations. To obtain acid-adapted cultures, one ml of each strain was added to 120 ml TSB with 1% (wt/vol) dextrose (TSB plus dextrose) incubated without agitation at 37°C for 18-24 h. To achieve nonadapted cultures, one ml of each strain was added to 120 ml TSB without dextrose (TSB-G, Difco Labs) and incubated without agitation at 37°C for 18-24 h. Each culture was centrifuged for 20 min at 2,500 x g, and the spent TSB was decanted and the bacterial pellet was re-suspended in 10 ml 0.1% peptone buffer (Bacto peptone, Difco Labs). The five strains of each bacterial type were pooled just prior to inoculation. Pooled inocula for each species was prepared by combining 3 ml of each strain. One ml of each pool was placed in 9 ml tubes containing 0.1% peptone and serially diluted to determine initial inoculum levels for each bacterial pool.
**Ground beef strip preparation.** Ground sirloin (98% fat free) was bought from a local grocery store. The bacterial cultures were prepared as previously described the same day as the meat was dehydrated. A commercial beef jerky spice mix (Original Flavor by American Harvest, Inc.; Chaska, MN) and cure mix were added to the meat at a ratio of one packet of spice mix and one packet cure mix per 454 g meat, according to the manufacturer’s instructions. Separate batches were inoculated with either 15 ml 0.1% peptone (control), 15 ml of acid-adapted bacterial strains, or 15 ml of nonadapted bacterial strains. To prevent possible cross-contamination, the control was mixed first, the nonadapted batch second and the acid-adapted third. Between the batches, the mixing bowl and mixer were cleaned with bleach, soap, and water. Each batch was mixed for 2 min at medium speed (model K5-A, Hobart Corporation, Troy, OH). Jerky strips (33.5 x 2.5 x 0.75 cm) were made using The Beef Jerky Works™ (BJW#1P, American Harvest, Inc.; Chaska, MN). Samples were taken during drying at time 0, 2, 4, 6, and 10 h.

**Whole beef strip preparation.** Bottom round steak was purchased from a local grocery store and used for the whole beef jerky strips. The edges of the beef were trimmed of visible fat and then sliced to approximately 9.0 x 1.5 x 1.5 cm strips. Strips were inoculated in a laminar air flow hood with 50 µl of each pool on separate one-third portions of each strip ensuring that the different pathogens did not overlap. The strips remained under the hood for 30 min to allow bacterial adhesion. The marinade (118.5 ml soy sauce, 29.6 ml Worcestershire sauce, 9.8 g garlic salt, 9.8 g black pepper and 9.8 ml liquid smoke/1820 g of meat) was added to the strips which were stored in ziplock bags at 4°C for overnight (16 h ± 2 h). The strips were blotted to remove visible liquid before
being placed in a 60°C pre-heated vertical air flow food dehydrator (Garden Master
Model # 1000, American Harvest, Inc.; Chaska, MN). Sampling for whole beef strips
was done at the following intervals: after inoculation, after marination (T:0), and during
drying at 4, 8, 12, 14 h.

**pH measurements.** pH was measured by the surface method using a surface electrode
and a Corning model 340 pH meter (Corning, Inc., Corning, NY).

**Microbial analysis and enrichment.** To determine initial microbial load on ground beef
strips, 25 g portions of beef were weighed, placed in a sterile stomacher bag with 225 ml
0.1% peptone buffer and pummeled (TekMar model 400; TekMar Co., Cincinnati, OH)
for 30 s on normal speed. To determine initial microbial load on whole jerky strips, one
strip was placed in a sterile stomacher bag with 225 ml 0.1% peptone buffer and
pummeled (TekMar model 400, TekMar Co; Cincinnati, OH) for 30 s on normal speed.

For both jerky types, serial dilutions of the samples were prepared using 0.1%
peptone. Portions of the stomached sample for both jerky types were spirally plated on
bismuth sulfite agar (BSA; Difco Labs), *Listeria* selective agar (LSA) (Oxoid;
Basingstoke, Hampshire, England), and sorbitol MacConkey agar (Oxoid) for
*Salmonella*, *Listeria* and *E. coli* O157:H7, respectively. The plates were incubated for 24
h at 37°C before colony forming units were counted, and the plates were re-incubated and
re-examined after an additional 24 h.

Subcultures of all pummeled samples in this study were enriched in the event that
the populations of the pathogens were reduced to levels not detectable (4.0 x 10^1) by
direct plating. Enrichment consisted of inoculating 1 ml from the stomacher bag into 9
ml portions of lactose broth (Difco Labs), UVM *Listeria* enrichment broth base (Oxoid), and modified tryptic soy broth (modified TSB; Difco Labs) (10 g casamino acids, 1.5 g bile salts No.3, 6.0 g dibasic, anhydrous sodium phosphate and 1.35 g potassium phosphate per liter of TSB) for *Salmonella*, *Listeria* and *E. coli* O157:H7, respectively. All three enrichment broths were incubated at 37°C for 18-24 h. After incubation, portions of the modified TSB cultures were streaked onto SMAC. Plates were incubated at 37°C for 24 h and examined for the presence of representative colonies. Subcultures were also made from the lactose broth into selenite cystine (Difco Labs) and TT broth Hajna (Difco Labs) and from UVM *Listeria* enrichment broth into Fraser broth (Difco Labs) and then incubated at 37°C for 24 h. After incubation of the broths, portions were streak plated onto bismuth sulfite agar (BSA; Difco Labs), XLD agar (Difco Labs) and brilliant green agar (BGA; Difco Labs) for possible *Salmonella* spp. isolates and onto LSA for possible *L. monocytogenes* isolates. Plates were incubated at 37°C for 24 h and examined for the presence of representative colonies.

Presumptive *Salmonella* spp. and *E. coli* O157:H7 isolates, from the enrichment steps were identified using the Micro-ID™ identification system for Enterobacteriaceae (Remel, Lenexa, KS) as per manufacturer’s instructions. Presumptive *Listeria* isolates were identified using the Micro-ID™ *Listeria* system (Remel, Lenexa, KS) as per manufacturer’s instructions.

**Dehydrators.** Two home-style vertical air flow food dehydrators (model # 1000, American Harvest, Inc.; Chaska, MN) were used to dehydrate both strip types. The dehydrators were preheated to 60°C prior to drying the strips. The internal air
temperature and internal temperature of an acid-adapted and nonadapted strip were monitored and recorded by a data recorder (model RD106 A, Omega, Stamford, CT) equipped with copper/constantan thermocouples (5TC-TT-T, Omega, Stamford, CT).

**Statistical analysis.** A split-plot type experimental design was used. Each experiment was replicated three times and the results analyzed using the GLM (general linear models) procedure in SAS (12). Acid-adapted and nonadapted treatments were compared separately for each organism (*E. coli*, *L. monocytogenes* and *Salmonella* spp.) for ground or whole beef strips. Plate counts were converted to log$_{10}$ counts and analyzed at each sampling time for significant differences (p < 0.05). Dehydrator number (1 or 2) and replication were main plot effects and subplot effects of treatment (acid-adapted or nonadapted) and interaction of dehydrator and treatment were also analyzed.

**RESULTS**

Analysis for *E. coli* and *Salmonella* spp. showed there was no significant difference (p< 0.05) in log reduction between the two dehydrators. There was also no interaction between dehydrator and treatment, for these two organisms. The dehydrator used significantly affected log reduction of *L. monocytogenes* (p<0.05) in whole strips at time 4 h, with dehydrator 2 resulting in a higher mean log reduction (4.33) as compared with dehydrator 1 (4.02) but this does not occur at the other sampling times. There was a significant interaction (p<0.05) between the dehydrator and treatment (acid-adapted and nonadapted) for whole strips inoculated with *L. monocytogenes* at sample time 4 h. For acid-adapted *E. coli* O157:H7 inoculated in ground beef, the maximum mean log reduction over the entire 10 h drying process was 6.22 as compared to 5.30 log reduction.
for nonadapted (Table 4.1). For whole jerky strips, this same trend resulting in a larger log reduction for acid-adapted (5.32) versus nonadapted *E. coli* O157:H7 (5.17) occurred at sampling time 8 h (Table 4.1). For both treatment types of *L. monocytogenes* and *Salmonella*, in ground beef strips, the populations achieved the largest log reduction at 10 h with nonadapted *L. monocytogenes* having a larger log reduction (4.51) than acid-adapted (4.28) and acid-adapted *Salmonella* resulting in a larger log reduction (4.73) than nonadapted (3.96) (Table 4.1). For whole jerky strips, *L. monocytogenes* had a mean log reduction of 4.94 as compared to nonadapted with a mean log reduction of 4.87. The log reduction was almost identical for whole strips inoculated with *Salmonella* (Table 4.1). The average pH for the acid-adapted and nonadapted cultures incubated overnight in broth was 4.88 and the nonadapted was 6.97, respectively.

**DISCUSSION**

Enterics thrive at pH 7.6-7.8 and as long as the pH is within 1 unit in either direction, pH homeostasis is responsible for survival of the cell (10). pH homeostasis is dependent on how permeable the bacterial cell membrane is to the protons in the acid. Whenever the cell’s pH homeostasis system cannot function to maintain a neutral pH, other systems within the cell are activated (9).

Acid tolerance response (ATR) protects log phase cells during long term exposure to low pH. Several steps are required to achieve ATR in a laboratory setting. Medium containing exponential phase cells is acidified to a moderate pH (near 5.5) for several h and then exposed to a lethal pH (< 4.0). This phenomenon may also result from bacteria being exposed to acidic conditions via acidic foods. Virulent bacterial strains exhibit
sustained ATR and this process can occur rapidly, within 20 min. ATR can be explained in part by the cell’s ability to repair damaged DNA caused by high H⁺ concentration.

The role of various intrinsic and extrinsic factors, strain, storage temperature, acidulent, and growth phase interact and influence the cell’s survival under adverse conditions (1, 8, 14). Acid-adaptation studies involving bacterial growth in media have been conducted (2, 3, 7, 14). Garren et al. (7) found that acid-adapted *E. coli* O157:H7 isolate 932 had a higher sodium chloride tolerance as compared to nonadapted and acid shocked cells. Polyphosphates and phosphates inhibit the acid response at pH 4.5-5.8 by competitively preventing protons form crossing the osmotic membrane. ATR is growth phase dependent and requires the stress-specific sigma factor rpoS for full induction (11). ATR broth studies involving enterohemorrhagic *E. coli* O157:H7 have shown the highest acid tolerance in late stationary phase. Similar patterns were seen in *Shigella flexneria* and *Salmonella* (1). In one study, cell survival was greater in apple juice than synthetic gastric fluid illustrating the necessity to study effects of food substrates on bacteria rather than in broth (14).

Several studies involving acid-adapted and nonadapted pathogens on beef jerky have been done recently and several of the studies reinforce the findings of the present research which concludes acid-adapted bacterial strains do not enhance survival of *L. monocytogenes* or *Salmonella*. Calicioglu et al. (6) examined the effect of storage on the proliferation of *Salmonella* on whole jerky strips that were inoculated after drying. The results showed that acid-adapted *Salmonella* decreased quicker than nonadapted cells for all treatments. The synergistic effects of modified marinades (e.g. dipping in Tween 20
or acetic acid for 10 min) and low water activity of the final product do provide
antimicrobial effects against post-contamination. A similar study examined the
inactivation of *L. monocytogenes* during drying and storage of whole beef strips. Again,
acid-adapted cells did not enhance survival. This study did conclude that modifications
in the marinade (e.g. dipping in Tween 20 or acetic acid for 10 min) did have an effect on
pathogen survival (4). However, another study by Calicioglu et al. (5) compared the
survival of acid-adapted and nonadapted *E. coli* O157:H7 on post-dried beef jerky and
found that acid-adapted *E. coli* O157:H7 enhanced inactivation during storage. While
this study did show that survival was better for acid-adapted *E. coli*, this experiment
inoculated the beef strips after the strips were dried so the effects of heat and other stress
factors encountered during drying did not factor into the inactivation of the cells.

The present experiment examined and compared population reduction of acid-
adapted and nonadapted *E. coli* O157:H7, *L. monocytogenes*, and *Salmonella* in two types
of beef jerky. The results showed that statistically there was no difference (p>0.05)
between the reductions of the populations of acid-adapted and nonadapted cells for *L.
monocytogenes* and *Salmonella* for both ground and whole beef jerky. There was a
significant difference (p= 0.0446) in the decrease of population types for ground strips
inoculated with *E. coli* O157:H7 unlike the whole strips which showed no difference.

The interaction and cross-protection of many systems within the bacteria enhance
survival (7, 9, 10, 13). For example, man attempts to keep this cross-protection to a
minimum by using hurdle technology in an attempt to inactivate and reduce pathogen
populations in a food. These results, like those of other similar studies, emphasize the
complexity of bacterial survival in adverse conditions and the need for more research with real food systems.
ACKNOWLEDGMENTS

This project was supported financially in part by a grant, validation of jerky processing and small-scale and home processors, funded by CSREES and USDA and by the Georgia Agricultural Experiment Stations. The authors would also like to thank Dr. Glenn Ware for his statistical expertise.
REFERENCES


   resistance response of enterohemorrhagic *Escherichia coli* in the presence of 

   survival and acid tolerance response of *Escherichia coli* O157:H7 and non-

   Destruction of acid-and nonadapted *Listeria monocytogenes* during drying and 

   *Escherichia coli* O157:H7 inoculated post drying on beef jerky treated with 

   modified marinades on survival of postdrying *Salmonella* contamination on beef 


Table 4.1. Mean log reductions between sample times for acid-adapted and nonadapted *E. coli* O157:H7, *L. monocytogenes* and *Salmonella* spp. for home-style ground and whole beef jerky strips.

<table>
<thead>
<tr>
<th>Time</th>
<th><em>E. coli</em> O157:H7</th>
<th><em>L. monocytogenes</em></th>
<th><em>Salmonella</em></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Acid-adapted</td>
<td>Nonadapted</td>
<td>Acid-adapted</td>
</tr>
<tr>
<td>Ground</td>
<td>2</td>
<td>1.08</td>
<td>1.42</td>
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<tr>
<td></td>
<td>4</td>
<td>4.79</td>
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<td>5.30&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
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<td>0&lt;sup&gt;c&lt;/sup&gt;</td>
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<td>1.18</td>
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<td>5.25</td>
<td>5.13</td>
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</tbody>
</table>

<sup>a,b</sup> Values with different letters are significantly different (p < 0.05). The p-values compared are between acid-adapted and nonadapted for each time interval and for each organism.

<sup>c</sup> Time 0 is after overnight marination at 4°C and before dehydration.
CONCLUSIONS

Jerky, solely relying on dehydration, may not be as safe as previously thought. Experimental data has shown that a heat step, in which the meat reaches an internal 71.1°C, is both safer from food pathogens and accepted by consumers. Salt plays an important safety and preservative role for jerky and reduced salt marinades may not produce a safe jerky product. The data from these experiments showed that bacterial cells, with the potential to acid adapt while being in the acidic marinade, do not have enhanced survival compared to nonadapted *E. coli, L. monocytogenes* and *Salmonella* spp.