

MICROBIAL TREATMENTS TO REDUCE OR ELIMINATE *CAMPYLOBACTER*
FROM BROILER CARCASSES, PARTS AND LIVERS

by

MELISSA ASHLEY LANDRUM

(Under the Direction of Nelson A. Cox and Jeanna L. Wilson)

ABSTRACT

The USDA's Food Safety and Inspection Service implemented stricter performance standards to reduce *Campylobacter* within poultry processing facilities. As the world's main cause of bacterial gastroenteritis, *Campylobacter* is commonly spread through poultry products and causes illness in approximately 850,000 people annually. The purpose of this research was to determine whether a low pH antimicrobial blend of sulfuric acid and copper sulfate, CMS PoultrypHresh™, effectively reduces *Campylobacter* prevalence in poultry, including whole carcasses, parts and livers. Four studies were performed: 1) naturally contaminated, whole carcasses treated with a 20 second, air agitated dip of water, PoultrypHresh™ or no treatment. Rinsates were plated directly and after enrichment; 2) skin-on split breasts and thighs inoculated with *Campylobacter* and treated with a 25 second, air agitated dip of PoultrypHresh™, water or no treatment; 3) inoculated poultry livers treated with a 15 second dip of PoultrypHresh™, PoultrypHresh™ with surfactant at 0.2% (PoultrypHresh™ Plus), water or no treatment; 4) inoculated skin-on thighs treated with sequential 6 second dips of PoultrypHresh™ and peracetic acid. Treatments were water dip followed by water dip,

PoultrypHresh™ by PoultrypHresh™, peracetic acid by peracetic acid, PoultrypHresh™ by peracetic acid, peracetic acid by PoultrypHresh™ or no treatment.

Results of this research demonstrated using PoultrypHresh™ is significantly effective on decreasing the prevalence of *Campylobacter* on broiler carcasses, parts and livers. The addition of surfactant enhanced the effectiveness of the chemical and further reduced *Campylobacter* on livers. Sequential dips exhibited interesting results. Peracetic acid dip followed by PoultrypHresh™ dip was significantly more effective than PoultrypHresh™ followed by peracetic acid. The sequence demonstrated significantly greater reductions than either peracetic acid or PoultrypHresh™ dipped consecutively. PoultrypHresh™ may be an effective intervention strategy for processing facilities to reduce *Campylobacter*. Sequential dipping may allow greater *Campylobacter* reductions and assist processors in meeting the strict performance standards.

INDEX WORDS: Antimicrobial, broiler, *Campylobacter*, CMS PoultrypHresh™, poultry processing, pathogen, foodborne illness, intervention strategy

MICROBIAL TREATMENTS TO REDUCE OR ELIMINATE *CAMPYLOBACTER*
FROM BROILER CARCASSES, PARTS AND LIVERS

by

MELISSA ASHLEY LANDRUM

BS, Georgia Southern University, Statesboro, Georgia, 2012

MS, University of Georgia, Athens, Georgia, 2015

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

DOCTOR OF PHILOSOPHY

ATHENS, GEORGIA

2018

© 2018

Melissa Ashley Landrum

All Rights Reserved

MICROBIAL TREATMENTS TO REDUCE OR ELIMINATE *CAMPYLOBACTER*
FROM BROILER CARCASSES, PARTS AND LIVERS

by

MELISSA ASHLEY LANDRUM

Major Professors: NELSON A. COX
JEANNA L. WILSON

Committee: BRIAN D. FAIRCHILD
MARK A. HARRISON
WOO K. KIM
MICHAEL P. LACY

Electronic Version Approved:

Suzanne Barbour
Dean of the Graduate School
The University of Georgia
December 2018

DEDICATION

This dissertation is dedicated to my mother, Mary Louise Landrum, father, Gerald Glenn Landrum, and the late Arthur Randy Cliett. With each roadblock life threw my way, these individuals remained by my side. My mother is the ultimate role model of strength and determination. She has sacrificed so much throughout my life to ensure I get to wherever my dreams are taking me. I will consider myself lucky to become even half the woman she is. She is more than a mother; she is my very best friend and inspiration. She has supported me in every way possible, been my number one fan, and never let me doubt myself. My father has helped encourage me and ensure I keep my ultimate goal in mind during the difficult times of graduate school. He pushed me to prove I could achieve whatever I wanted in life, not only to others, but also to myself. He was sure to let me know how proud he was of the woman I have become. Randy was an immensely special person in my life, who I miss every single day. He taught me the meaning of hard work and to put my heart into everything I do. It was through time with him I learned how strong of a person I truly am and what I am capable of. He made me tough, thick skinned, determined, and even more hardheaded than I already was. These people made me the woman I am today. I would not trade a single memory for anything and I will be forever grateful to the three of them for all they have done. I only hope I can continue making them proud.

ACKNOWLEDGEMENTS

I would like to acknowledge Dr. Nelson Cox for his support and assistance as not only a major professor, but also a mentor and a friend. Dr. Cox has taught me so much during my time with him and I appreciate it more than he will ever know. He has always stood by me, been understanding and helpful, and goes out of his way to support me. I would also like to acknowledge Dr. Jeanna Wilson for her support and assistance throughout my doctorate program. She has kept me on track and made sure I had everything I needed to reach my goal. I owe her a very special thank you and I truly do appreciate everything. I would also like to acknowledge the late Dr. Scott Russell. He took a chance on me, pushed me to be all I could, and made sure I was taken care of when he knew he would no longer be there. He will forever be a huge part of my great journey and never be forgotten.

TABLE OF CONTENTS

	Page
ACKNOWLEDGEMENTS	v
LIST OF TABLES	ix
CHAPTER	
1 INTRODUCTION	1
Purpose of the Study	1
How This Study is Original	2
Expected Results	3
References	3
2 LITERATURE REVIEW	6
Overview of the Commercial Broiler Industry	7
Poultry Demand Worldwide	8
Governmental Agencies for Food Safety	9
Microorganisms of Concern in Poultry	12
Pathogen Hazard Points in Processing	15
Pathogen Testing Strategies	24
The Addition of Processing Aids	29
<i>Campylobacter</i>	43
References	53

3	LOW PH PROCESSING AID TO LOWER THE PRESENCE OF NATURALLY OCCURRING <i>CAMPYLOBACTER</i> ON WHOLE BROILER CARCASSES.....	89
	Abstract.....	90
	Introduction.....	90
	Materials and Methods.....	92
	Results and Discussion	94
	References.....	95
4	TREATMENT WITH A LOW PH PROCESSING AID TO REDUCE <i>CAMPYLOBACTER</i> COUNTS ON BROILER PARTS	98
	Abstract.....	99
	Introduction.....	100
	Materials and Methods.....	101
	Results and Discussion	103
	References.....	106
5	REDUCTION OF <i>CAMPYLOBACTER</i> ON CHICKEN LIVERS USING A LOW ACID PROCESSING AID	109
	Abstract.....	110
	Introduction.....	110
	Materials and Methods.....	113
	Results.....	114
	Discussion.....	115
	References.....	117

6	REDUCTION OF <i>CAMPYLOBACTER</i> ON POULTRY THIGHS USING SEQUENTIAL TREATMENTS OF ANTIMICROBIALS	121
	Abstract	122
	Introduction.....	123
	Materials and Methods.....	124
	Results.....	126
	Discussion.....	127
	Conclusions.....	129
	References.....	129
7	CONCLUSIONS.....	134
	References.....	137
8	REFERENCES	141

LIST OF TABLES

	Page
Table 3.1. Number of direct, enriched, and overall samples positive for <i>Campylobacter</i> in untreated, water treated, and PoultrypHresh™ treated at pH 1.4 for 6 replications. Samples not positive on direct analysis were enriched and re-evaluated	97
Table 4.1. Average Log ₁₀ cfu/mL of <i>Campylobacter coli</i> recovered from parts dip treated with no treatment, water, or PoultrypHresh™ for 25 seconds with agitation in 3 study replications	108
Table 5.1. Average Log ₁₀ cfu/mL of <i>Campylobacter coli</i> recovered from livers dip treated with no treatment, water, PoultrypHresh™, or PoultrypHresh™ Plus for 15 seconds with no agitation in 3 study replications	120
Table 6.1. Average Log ₁₀ cfu/mL of <i>Campylobacter coli</i> recovered by replication from sequentially dip treated thighs with no treatment, water - water, PoultrypHresh™ - PoultrypHresh™, Peracetic acid - Peracetic acid, PoultrypHresh™ - Peracetic acid, or Peracetic acid - PoultrypHresh™ in 5 study replications.....	133

CHAPTER 1

INTRODUCTION

Purpose Of This Study

The purpose of this study is to evaluate new treatment methods for reducing *Campylobacter* in poultry processing operations. *Campylobacter* is a serious pathogen and one of the leading causes of bacterial foodborne diarrheal illness in the United States (Bashor et al., 2004; USDA, 2013). It causes 850,000 illnesses annually in the U.S., including 8,000 hospitalizations and 80 deaths (USDA, 2017). Poultry and poultry products were found to be the most common host, as 50 – 70% of *Campylobacter* cases were shown to be associated with poultry (Tauxe, 1992; Allos, 2001; Humphrey et al., 2007). As *Campylobacter* species continue developing antibiotic resistance to a variety of commonly used drugs, it becomes an even more dangerous pathogen for humans.

The stricter performance standards implemented by FSIS now allow only a maximum of 15.7% *Campylobacter* on whole carcasses and 7.7% on parts (Inspection Methods, 2018). These standards make it essential for processors to reduce *Campylobacter* levels, as processing plants can be shut down by FSIS if standards are not met. Many antimicrobials have been evaluated for *Campylobacter* reduction and have shown varied results. This research evaluates the efficacy of a new antimicrobial blend of sulfuric acid and copper sulfate, PoultrypHresh™, administered by dip treatment on broiler carcasses, parts and livers to reduce *Campylobacter*. A new method of treatment is

also evaluated using consecutive acid dips in different sequences to determine if the efficacy of chemicals is altered.

How This Study Is Original

There is a tremendous amount of research evaluating a variety of chemicals for treatment at various points of poultry processing. The antimicrobial evaluated in these studies was not previously published in literature for effectiveness in any type of poultry treatment. This testing covers first and secondary processing applications on whole carcasses, parts and livers, all known to have high prevalence of *Campylobacter*. PoultryHresh™ was developed by the company, CMS Technology, Inc., as an antimicrobial, liquid additive made from GRAS ingredients and applied directly to food products to eliminate an assortment of foodborne pathogens.

Stopforth et al. (2007) determined individual intervention points would not significantly reduce bacterial prevalence on finished carcasses and multiple points are necessary for significant reductions. Researchers determined a multi-hurdle approach is necessary for intervention strategies (Leistner and Gorris, 1995; Bacon et al., 2000). No previously published literature was found on sequential or consecutive dip treatments or how they may alter the level of bacterial pathogens. This research evaluated consecutive dips of the same chemical, as well as sequential dips of different chemical antimicrobials at a short treatment time of 6 seconds to evaluate whether differences in efficacy were demonstrated. Short dip treatments are practical for the fast pace of the modern broiler processing facility.

Expected Results

Since CMS PoultryHresh™ is a relatively new antimicrobial aimed at reducing the prevalence of pathogens, there are no specific expectations as to the level, if any, of reduction will be found. It seems more likely greater reductions would be demonstrated on livers treated, rather than whole carcasses or parts, as broiler skin is known to harbor bacteria. Bacteria can become entrapped and proliferate in the skin's superficial layers (Thomas and McMeekin, 1980; Lillard, 1989; FAO, 2009). Researchers have shown *Campylobacter* growth is inhibited at a pH below 4.9 or above 9.0 and rapid death of *Campylobacter* occurs below pH 4.0 (NZFS, 2001). PoultryHresh™ is a low acid antimicrobial, well below pH 4.0, and is therefore likely to show some level of reduction in *Campylobacter* prevalence. There are no specific expectations for the results of sequential dip treatments, as this method has not been present in published literature. Research has shown multiple intervention points more likely to reduce pathogens (Leister, 1995; Bacon et al., 2000; Stopforth et al., 2007) throughout processing, therefore it is possible consecutive chemical dips or different chemicals in a sequence may lead to greater reductions in *Campylobacter* prevalence.

References

- Allos, B. 2001. *Campylobacter jejuni* infections: update on emerging issues and trends. Clin. Infect. Dis. 32(8):1201-1206.
- Bacon, R. T., K. E. Belk, J. N. Sofos, R. P. Clayton, J. O. Reagan, and G. C. Smith.

2000. Microbial populations on animal hides and beef carcasses at different stages of slaughter in plants employing multiple-sequential interventions for decontamination. *J. Food Prot.* 63(8):1080-1086.
- Bashor, M. P., P. A. Curtis, K. M. Keener, B. W. Sheldon, S. Kathariou, and J. A. Osborne. 2004. Effects of carcass washers on *Campylobacter* contamination in large broiler processing plants. *Poult. Sci.* 83:1232-1239.
- FAO [Food and Agriculture Organization of the United Nations]. 2009. *Salmonella* and *Campylobacter* in chicken meat: meeting report. Page 56 in Microbiological Risk Assessment Series No. 19. Rome.
- Humphrey, T., S. O'Brien, and M. Madsen. 2007. Campylobacters as zoonotic pathogens: a food production perspective. *Int. J. Food Microbiol.* 117:237-257.
- Inspection Methods. 2018. Pathogen reduction – *Salmonella* and *Campylobacter* performance standards verification testing. Food Safety Inspection Service (FSIS). <https://www.fsis.usda.gov/>.
- Leistner, L. 1995. Food preservation by hurdle technology. *Trends Food Sci. Technol.* 6:41-46.
- Lillard, H. S. 1989. Incidence and recovery of salmonellae and other bacteria from commercially processed poultry carcasses at selected pre- and post-evisceration steps. *J. Food Prot.* 52:88-91.
- New Zealand Food Safety (NZFS). 2001. *Campylobacter*. <https://www.foodsafety.govt.nz/elibrary/industry/Campylobacter-Organism-Causes.pdf>.
- Stopforth, J. D., R. O'Connor, M. Lopes, B. Kottapalli, W. E. Hill, and M. Samadpour.

2007. Validation of individual and multiple-sequential interventions for reduction of microbial populations during processing of poultry carcasses and parts. *J. Food Prot.* 70(6):1393-1401.

Tauxe, R. V. 1992. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. Pages 9-19 *Campylobacter jejuni: current status and future trends*. I Nachamkin, M. J. Blaser, and L. S. Tompkins, eds. American Society for Microbiology. Washington, D.C.

Thomas, C. J., and T. A. McMeekin. 1980. Contamination of broiler carcass skin during commercial processing procedures: an electron microscopic study. *Appl. Environ. Microbiol.* 40:133-144.

United States Department of Agriculture (USDA). 2013. Food Safety Education – *Campylobacter* Question and Answers. <http://www.fsis.usda.gov/>.

United States Department of Agriculture (USDA). 2017. Cost estimates of foodborne illnesses. <https://www.ers.usda.gov/data-products/cost-estimates-of-foodborne-illnesses/>.

CHAPTER 2
LITERATURE REVIEW

Overview of the Commercial Broiler Industry

Industry Evolution

Throughout the last century, the commercial broiler industry has expanded as consumer demand continues to rise. Since 1960, poultry production has increased close to 900% (USDA, 2016a), with over 55.5 billion pounds produced in 2017 (USDA, 2017a). The tremendous growth of the industry is attributed to many improvements such as increased availability, convenience, lower prices, vertical integration, technological advancements, and genetic improvements. In the past, poultry was mainly served on special occasions, but the industry's progression allowed poultry to surpass consumption rates of all other meat industries by 1985 (Havenstein et al., 2006). The development of vertical integration, allows a single firm, or integrator, to control all aspects of production, processing, and overall product cost. This increased product distribution allowed the production process to run more efficiently (Anthony, 1998; Ferrara, 2005; Havenstein et al., 2006).

Further processing poultry refers to the conversion of raw carcasses into more convenient, cut up forms, which may have the addition of breading, batter, or were altered for added value (Baker and Bruce, 1995). The convenience and value added when poultry is further processed was a major contributor to the industry's rapid growth. While it was customary to purchase whole carcasses in the past, the industry began allowing consumers to choose desired parts. This appealed to consumers increasingly busier lifestyles, therefore further processed and ready-to-eat products continued to gain popularity (Anthony, 1998). Consumers were also attracted to poultry for its seemingly greater health benefits compared to other meat industries. Poultry is known to be lower in

fat, sodium, and cholesterol compared to other types of meat (Petracci and Cavani, 2012). It was reported in 1976, by the Royal College of Physicians and the British Cardiac Society, that limiting red meat and increasing poultry consumption is beneficial to consumer health (Wang et al., 2010). Petracci et al. (2014) determined fat content differs due to the location lipids accumulate in the animal. Red meat has intramuscular fat, known as marbling, which is difficult to separate from the muscle (Petracci et al., 2014). Lipid content in poultry occurs directly under the skin, which easily detaches from the meat and makes it lower in fat (Sams and Alvarado, 2010; Petracci et al., 2014). This gives poultry an edge over other meat industries, as healthy living continues to gain popularity in today's society.

Poultry Demand Worldwide

The U.S.'s evolution of vertical integration currently includes approximately 35 large companies. These companies oversee the growing, processing, and marketing of their own birds, allowing them to ensure quality is maintained at all aspects (NCC, 2018). Companies outsource production contracts to family farms, where 95% of U.S. broiler chickens are raised (NCC, 2018). This accounts for approximately 25,000 family farms, while only 5% of grow-out locations are company owned (NCC, 2018). The top five states for poultry production are Georgia, Arkansas, Alabama, North Carolina, and Mississippi (NCC, 2018). Overall, the United States has the largest broiler industry in the world and exported 19% of production to other countries in 2015 (NCC, 2018). Top export countries include Mexico, Canada, and Hong Kong.

Americans purchase more poultry products than any other country in the world, as poultry has risen to the number one protein source. The industry produced almost 9 billion broilers, accounting for 55.5 billion pounds of live weight in 2017 (USDA, 2017a). More than 40 billion pounds of this was marketed and sold as ready-to-eat (cooked) product (NCC, 2018). Currently, the U.S. has 180 processing facilities, employing over 280,000 people and indirectly employing more than 1.3 million (NCC, 2018). In 1960, per capita poultry consumption averaged 23.6 pounds, which increased to over 90 pounds by 2017 (NCC, 2018).

Governmental Agencies for Food Safety

The two agencies responsible for overseeing the safety of food in the United States are the United States Department of Agriculture's Food Safety and Inspection Service (USDA-FSIS) and the Food and Drug Administration (FDA). Meat, poultry and eggs are regulated by the USDA, while the FDA oversees the rest of the food supply and ingredients. In addition, each state has its own agencies overseeing food production, which vary in complexity and strictness. The food industry in the U.S. employs 14 million individuals and makes up 20% of the U.S.'s gross national product (CIA World Factbook, 2017).

United States Department of Agriculture (USDA)

In 1862, Abraham Lincoln founded the USDA by signing an act of congress into law (Rasmussen, 1986). Agriculture in the U.S. has continuously progressed since, as the population and overall demand for food have risen. The USDA is now made up of 29

agencies with nearly 100,000 employees working in over 6,000 locations (USDA, 2016b). The department's mission statement is to "provide leadership on food, agriculture, natural resources, rural development, nutrition, and related issues based on sound public policy, the best available science, and efficient management" (USDA, 2018). The USDA oversees farm management and operates an audit and certification program to verify each farm follows good agricultural practices.

Food Safety Inspection Service (FSIS)

A USDA agency, FSIS, is responsible for overseeing meat and poultry processing plants. FSIS inspects products traded or sold across state lines, but a processing facility only selling in state, is only required to have state inspectors (Keenan et al., 2015). Processed poultry staying in state accounts for 16% of the U.S.'s poultry production (USDA, 1984). FSIS inspectors visually inspect every carcass undergoing slaughter operations, including more than 8 billion broilers and 125 million livestock in several thousand processing facilities each year (Keenan et al., 2015). FSIS only oversees processing procedures and are not mandated to inspect farms. In 1957, the Poultry Inspection Act was passed requiring all birds to be inspected prior to and after slaughter (USDA, 1984). Other requirements for this act include inspecting facilities to maintain sanitary conditions, slaughtering and processing operations, verification techniques, labeling accuracy, and imported poultry products (USDA, 1984).

FSIS focuses on ensuring processing procedures are monitored and performance standards set to evaluate the effectiveness of antimicrobials being used in each facility. In 1996, the USDA-FSIS implemented the Hazard Analysis and Critical Control Point

Program (HACCP) to be used in all meat and poultry processing facilities to reduce hazards and produce safer products (Federal Register, 1996; PEW, 2016). HACCP will be discussed in detail later in this literature review. Foodborne pathogen prevalence remained high in processing facilities causing the USDA-FSIS to introduce stricter performance standards for each facility focused on controlling *Salmonella* and *Campylobacter* in poultry products (USDA-FSIS, 2015). New standards mandate *Salmonella* prevalence must remain below 9.8% (5 of 51 samples) in broiler carcasses and 15.4% (8 of 52) in broiler parts (Inspection Methods, 2018). *Campylobacter* levels for carcasses must remain below 15.7% (8 of 51) and 7.7% (4 of 52) in poultry parts (Inspection Methods, 2018). The new standards incentivize processing facilities to establish better techniques for reducing and eliminating bacteria throughout processing procedures.

Food and Drug Administration (FDA)

The FDA is a federal agency within the U.S. Department of Health and Human Services and is one of the oldest consumer protection agencies in the U.S., employing over 9,000 people (FDA, 2015). There are approximately 1,100 inspectors based in 157 cities, who visit more than 15,000 factories and plants annually to inspect and ensure processors are accurately labeling and following required health laws (FDA, 2015). Missions of the FDA are based on protecting and promoting the health of humans and animals. This includes ensuring safe food and animal feeds, effective animal drugs, science-based standards for evaluating foodborne illnesses, protection from intentional contamination and ensuring reliable label information (Keenan et al., 2015). The FDA is

responsible for 80% of the U.S.'s food supply, with the exception of meat and poultry products. The agency enforces regulations and conducts inspections of manufacturing facilities and processing plants. The FDA works with other agencies, such as state and local officials, and funds grants for state inspections and building of new infrastructures (Keenan et al., 2015). The FDA provides guidance, training programs, technical assistance, and scientific advice to state and local agencies, as well as the industries they regulate. FDA samples are evaluated for contaminants, such as pesticides and bacteria, making corrective actions and occasionally recalls, if necessary. In January 2011, the Food Safety Modernization Act (FSMA) was signed, aimed at shifting focus to prevention of food contamination in products, rather than only responding to it when found (Keenan et al., 2015).

Microorganisms of Concern in Poultry

Poultry is the most common vehicle for transmission of foodborne microorganisms to infect and sicken consumers (FAO, 2009). One of the common methods of contamination is from consuming raw or undercooked chicken (USDA-FSIS, 2014). The majority of foodborne illnesses result from contamination by improper handling during preparation and cooking (Mead et al., 1999; Nauta et al., 2009; USDA-FSIS, 2014). When handling raw product, consumers should be informed of proper handling techniques to prevent cross contamination from occurring (USDA-FSIS, 2014).

While foodborne pathogens are a major concern, researchers also focus on spoilage bacteria, such as *Pseudomonas* and *Achromobacter* (Davies and Board, 1998). Undercooked or mishandled products may also harbor spoilage organisms, with the

potential to greatly reduce shelf life (Centers for Disease Control and Prevention, 1993). Psychrotrophic microorganisms that remain on the surface after processing procedures can multiply and quickly spoil fresh products (Gallo et al., 1988; Russell et al., 1996; Hinton et al., 2002). Many factors determine how quickly poultry spoils including storage temperatures, packaging types, and the number or type of psychrotrophic bacteria present (ICMSF, 1998).

Common Foodborne Pathogens and Illnesses

Approximately 76 million Americans are affected by food poisoning annually. More than 2 million of these illnesses occur from consumption of meat products and cost the U.S. more than \$5.7 billion dollars a year (Batz et al., 2012). The U.S. Centers for Disease Control and Prevention (CDC) reported meat and poultry account for approximately 40% of all bacterial foodborne illnesses within the country (Painter et al., 2013). The CDC estimated 1 in 6 Americans become sick from food products annually, leading to 128,000 hospitalizations and 3,000 deaths (Scallan et al., 2011a). Thirty-one pathogens are found responsible for the 9.4 million foodborne illness cases reported and 90% of these were attributed to only five pathogens: norovirus, *Salmonella*, *Clostridium perfringens*, *Campylobacter*, and *Staphylococcus aureus* (Scallan et al., 2011b). The five pathogens causing 88% of hospitalizations are *Salmonella*, norovirus, *Campylobacter*, *Toxoplasma gondii*, and Shiga toxin-producing *E. coli* (Scallan et al., 2011b). Approximately, 88% of deaths were caused by *Salmonella*, *T. gondii*, *Listeria monocytogenes*, norovirus or *Campylobacter* (Scallan et al., 2011b). Poultry products

remain the most common vehicle of transmission for these pathogens, with the exception of norovirus and *Staphylococcus aureus* (Batz et al., 2012).

Challenges with Foodborne Pathogens

Foodborne pathogens, such as *Salmonella*, have the ability to adhere or attach to poultry skin, even after undergoing adequate rinsing procedures (Thomas and McMeekin, 1982; Lillard, 1988; Lillard, 1989). Broilers entering the processing facility with dirt and feces covering their feathers are likely to have high bacterial loads, making it seemingly inevitable for processing equipment, additional carcasses, or final product to be contaminated (Cason and Hinton, 2006). Visually dirty feathers have bacteria either adhered to the skin's surface or entrapped within (Lillard, 1989; Kim et al., 1993; Allen et al., 2003). The likelihood of bacterial attachment was correlated to exposure time and reported to happen within 15 seconds (Lillard, 1985). Other scientists found no time dependence, but instead related attachment to the level of bacteria present (McMeekin and Thomas, 1978). Connor and Biligli (1994) found no relationship between exposure time, bacteria level, or temperature during attachment. These conflicting results demonstrate bacterial attachment may be complex and further research is required for better understanding.

Foodborne pathogens are zoonotic and therefore, able to be transferred between animals and humans. This property allows them to be introduced at any point of poultry production, from day-of-hatch chicks, to the day of slaughter, and even up until food is consumed. Microorganisms are constantly evolving and changes in genetic structure can make a pathogen more efficient and able to survive new or more difficult environments

(PEW, 2016). Once pathogens become resistant to antimicrobial drugs or treatments, they may re-emerge with enhanced virulence, making them more difficult to be controlled, or acquire the ability to inhabit new hosts (PEW, 2016). Subtypes of existing pathogens that became antimicrobial resistant are responsible for a number of outbreaks, becoming a concern to consumer health (DeWaal et al., 2012). When a completely new pathogen emerges, it can be an exceedingly dangerous threat due to unknown factors such as epidemiology, transmission routes, preventable vaccines or treatments (PEW, 2016).

Pathogen Hazard Points in Processing

HACCP

Processing procedures provide many opportunities for microbial contamination to occur. Facilities must establish proper intervention points at such locations to assist in reducing or eliminating likelihood of pathogen cross contamination. The Hazard Analysis and Critical Control Point Program (HACCP) was developed by a company called Pillsbury, who was attempting to provide safer foods for NASA astronauts in 1959 (Northcutt and Russell, 2010). In 1996, the USDA-FSIS imposed the HACCP program to be used in all meat and poultry plants, aimed at reducing hazards associated with meat and poultry (Federal Register, 1996). Goals were to reduce or eliminate foodborne illnesses by mandating the industry to monitor the potential hazard points throughout processing (Dreyfuss et al., 2007; Mbata, 2017). Hazards were defined as either biological, physical or chemical reasons a product would become unsafe for consumption (Northcutt and Russell, 2010). The CDC consider HACCP an important tool in decreasing foodborne illnesses (Centers for Disease Control and Prevention, 2002; Vugia

et al., 2003). The National Advisory Committee on Microbiological Criteria for Foods (NACMCF) supported evaluation of the entire process and determined testing only end products was not effective for monitoring food safety. Rather, testing throughout the entire process evaluates all potential points of microbial contamination and proper processing specifications. Adding preventive measures at these processing locations provides a better means to ensure foodborne pathogens lesser opportunity for infection or contamination (Fortin, 2003).

Each company should first establish a HACCP team, including individuals from all parts of production. Ideally, these individuals would cover all areas of expertise, to enable well-informed decisions to be made in regard to HACCP plans. Teams would then write a HACCP plan for each product the plant produces, including information on how the product is used, packaging materials, shelf-life, temperature, location sold, labeling instructions, and any special product information (Northcutt and Russell, 2010). All plant employees should undergo training in HACCP concepts to understand the overall goal and assist in properly following the plans.

When designing a plan, each product requires an analysis of hazards to evaluate all aspects of that product's production. Any biological, chemical or physical risks should be listed, and a preventative action developed to address each (Northcutt and Russell, 2010). HACCP teams should then determine critical control points (CCP's) where these risks are likely to occur. A CCP decision tree may be used to identify whether specific points are actually true CCP's. Critical limits will then be set for each point, allocating the appropriate amount of variation allowed before safety becomes an issue (Northcutt and Russell, 2010). These critical limits are not meant to assist in product quality, but

only be related to safety applications. It is essential for each CCP to be monitored, ensuring critical limits are maintained. Monitoring systems should be simple and allow quick assessment of the risks.

Action plans should be made for each CCP and employees prepared on proper handling for situations where CCP's are not being met (Northcutt and Russell, 2010). Each action plan should determine potential problems, including who addresses the issue, how to fix it, what to do with product produced during the risk period, and proper recall instructions (Northcutt and Russell, 2010). Detailed records are mandatory and required for all CCP's. They must include responsibilities, dates, data, flow diagrams, critical limits, preventative measures, action and procedures, packaging information, and check sheets for employee monitoring (Northcutt and Russell, 2010). HACCP plans should be evaluated regularly and altered whenever any changes occur within a plant. These changes may include new equipment, updated products, new products, or altered handling procedures. When HACCP plans are being followed correctly and employees well educated on proper techniques, HACCP can play a role in reducing bacterial foodborne illnesses (Centers for Disease Control and Prevention, 2002; Vugia et al., 2003).

Transportation

When birds are transported from a grow out farm to a processing facility, cross-contamination between birds is very likely. Broilers shed *Salmonella* and *Campylobacter* in their feces when undergoing stress, greatly increasing pathogen levels (FAO, 2009). *Campylobacter* prevalence can increase 100 times between cooping of broilers and entry

into processing (Berrang et al., 2018). Birds leaving the farm with high levels of dirt and feces attached to their feathers were shown to have high bacterial loads internally and externally. During processing, this contamination is spread to equipment, other carcasses, and possibly final products. Lillard (1989) determined broilers arriving to processing facilities may have bacteria firmly attached or entrapped within their skin, also increasing likelihood cross contamination occurs during processing.

One method for reducing cross-contamination is ensuring adequate feed withdrawal times when loading birds for transport. The ideal feed withdrawal period is 8 - 12 hours of feed restriction, which should begin when access to feed is restricted at the farm and include catching, transport and holding prior to slaughter (Northcutt et al., 2003b). If given sufficient time, the level of fecal material and ingesta within the bird is reduced and likelihood of contamination during processing lowered (FAO, 2009). If intestines remain filled with feces, it significantly increases the chances they will be nicked or ruptured during processing, resulting in cross contamination. If birds are withdrawn from feed for more than 12 hours, intestines will become weak and easily torn by plant machinery. Since high stress levels increase *Salmonella* and *Campylobacter* shedding in feces, it is important to attempt minimal stress during transport. Some ways to reduce stress are giving birds adequate space and protection from environmental conditions such as temperature, humidity, and pressure fluctuations (FAO, 2009). Once reaching the processing facility, birds should be given shelter during holding for protection from extreme weather conditions (FAO, 2009). Another way to minimize cross-contamination is by washing and sanitizing transport coops, although this process is not always implemented at facilities (FAO, 2009).

Scalding

Scalding is meant to open feather follicles, making them easier to remove within the picker. Follicles remain open throughout the process until birds enter the cold chiller water, which causes follicles to close. Researchers have shown the scalding to be a common point of cross contamination (Bailey et al., 1987). One reason may be that scalding and picking remove the epidermis layer of the skin, providing new surfaces for bacterial colonization throughout the remainder of processing procedures (Thomas and McMeekin, 1980). Scalders running efficiently can reduce cross contamination, for example the temperature of scalding water for reducing bacterial growth and presence on the skin's surface (Mulder et al., 1978; Geornaras et al., 1995; Rouger et al., 2017). It was demonstrated, however, contamination level correlates to bacterial concentration present within processing water (McMeekin and Thomas, 1978). Bacteria can migrate from contaminated water and attach to skin or become entrapped in skin's crevices or ridges (Thomas and McMeekin, 1980; Lillard, 1988). Scalders with inadequate water flow can accumulate dirt and feces, causing extremely high levels of bacterial contamination (Hinton et al., 2004a; Hinton et al., 2004b; Cason and Hinton, 2006; USDA-FSIS, 2008).

To reduce likelihood of cross contamination, scalders should be counter-current in flow and have adequate fresh water added (FAO, 2009). Some chemicals, such as sodium chloride, sodium bisulfate, peracetic acid, or acetic acid (USDA-FSIS, 2018) are approved for reducing cross contamination in the scalding, aimed especially at pathogenic organisms such as *Campylobacter* and *Salmonella* (Mulder et al., 1978; Cason et al., 2000; Hinton et al., 2004b; Russell, 2007). An efficient scalding can act as a washing

machine and remove some contamination on birds entering with high quantities of dirt and feces on their feathers, although it greatly depends on scald type and operating conditions (Hinton et al., 2004a; Hinton et al., 2004b; Cason and Hinton, 2006; USDA-FSIS, 2008).

Feather Picking

Researchers have found the feather picker to be one of the main points for cross-contamination to occur during processing (Lillard, 1986; Hinton et al., 2004c; Arnold, 2007; Arnold and Yates, 2009; Veluz et al., 2012). Picker fingers are designed to remove feathers and are made from a rubber material shown to resist bacterial attachment compared to other materials (Arnold, 2007). Picker fingers should be monitored closely to ensure they do not become worn, cracked, or soiled from extensive use. This extensive use allows greater likelihood for bacterial attachment or formation of a biofilm (Kim et al., 1993; Arnold and Silvers, 2000). If a flock has non-uniform birds of various sizes, the picker may damage the skin during feather removal (Thomas and McMeekin, 1980; FAO, 2009). This allows bacteria an opportunity to become lodged and proliferate in the superficial layers of the skin (Thomas and McMeekin, 1980; FAO, 2009). Once fingers are contaminated, they have the potential to spread pathogens to any subsequent carcasses entering the picker (Berrang et al., 2001).

Research evaluating *Campylobacter* level on carcasses prior to entering the picker compared to exiting, found a 3.70 log cfu/mL increase as exiting carcasses (Berrang and Dickens, 2000). Other studies showed similar results, indicating cross-contamination is highly likely in the picker (Acuff et al., 1986; Izat et al., 1988). Researchers found high

Campylobacter levels (94.4%) in water samples taken from the picker, also indicating the high likelihood for cross contamination of birds entering the picker throughout the day (Wempe et al., 1983).

Evisceration

Evisceration is removal of the digestive tract from the carcass, which holds an array of bacterial colonies. It is essential for this to be completed without nicking or spilling the intestinal contents (Hue et al., 2011; Pacholewicz et al., 2016). The gastrointestinal tract can harbor pathogenic bacteria, such as *Campylobacter* and *Salmonella*, which have been shown to be correlated to the level of contamination on the exterior of carcasses and number of pathogens in the ceca (Hue et al., 2011; Pacholewicz et al., 2016). When intestinal contents are spilled, the likelihood of contamination and cross contamination rises substantially (Berrang et al., 2001). When evisceration is complete, bacterial populations are significantly reduced, possibly from the removal of paws and the spray wash following evisceration (Geornaras and von Holy, 2000).

The most important factor in proper evisceration is feed withdrawal of birds prior to entering the processing facility. The process begins at the farm when access to feed is restricted at the appropriate time and the remainder of time is properly managed in transport and holding by the processing facility. Evisceration machinery should be set for the size of the birds and uniformity reduces the chance of intestinal tears. Chemical dips and sprays may be a beneficial addition at this point and reduce carcass bacteria. Inspectors must evaluate carcasses closely for fecal contamination and immediately

remove the carcass from the processing line if found. These carcasses would undergo reprocessing procedures, which are essential to reducing contamination.

Air Versus Immersion Chilling

The chiller is essential to poultry processing and can affect overall meat quality and the growth of microorganisms (Petрак et al., 1999; McKee, 2001; Sanchez et al., 2002). Chilling also affects water holding capacity, flavor, appearance, microbial flora, and shelf life (Davies and Board, 1998; Schreurs, 2000; Fletcher, 2002). Carroll and Alvarado (2008) determined chilling can potentially prevent pathogen growth, increase shelf life and provide safer products for consumers. The two common chilling methods are immersion and air, each having advantages and disadvantages. Immersion chilling is considered more efficient and is commonly used in the U.S., while Europe, Brazil and Canada prefer air chilling (Sams, 2001; Fluckey et al., 2003; Carciofi and Laurindo, 2007; Huezo et al., 2007; Berrang et al., 2008; Carroll and Alvarado, 2008).

The purpose of chilling is to reduce carcass temperature within 4 hours of the bird being slaughtered. Rapid chilling limits growth of spoilage and pathogenic bacteria and helps to improve shelf life. Immersion chilling requires carcasses to be submerged in cold water (0 - 4°C) until deep muscle carcass temperature is 4°C. Research has demonstrated the potential of immersion chilling to reduce microbial contamination when certain requirements are met. Water flow must be countercurrent, contain agitators and be chlorinated (Dickens and Whittemore, 1995; Petрак et al., 1999; Bilgili et al., 2002). Chlorine has been a common antimicrobial used in chillers since the 1950's (50 ppm) and has shown to improve carcass quality and shelf life (Drewniak et al., 1954).

Countercurrent flow maximizes chilling by causing a temperature gradient, but leads to some water retention (USDA, 2001).

Many studies have evaluated whether immersion chilling leads to cross-contamination since birds come in contact with each other (Bailey et al., 1987; Mead et al., 1995; Bilgili et al., 2002; Whyte et al., 2007). Researchers have demonstrated immersion chilling can reduce total aerobic bacteria, coliforms, *Escherichia coli*, *Salmonella* and *Campylobacter* on poultry carcasses (Berrang and Dickens, 2000; Bilgili et al., 2002; Northcutt et al., 2003a; Northcutt et al., 2008). Demirok et al. (2013) showed immersing carcasses in chlorinated water reduces *Campylobacter* by 43% and *Salmonella* by 39.7%. Oyarzabal et al. (2004) demonstrated greater prevalence of *Campylobacter* positive carcasses once samples were enriched, suggesting immersion chilling is unable to adequately reduce *Campylobacter* and it remains on carcasses post-chill.

When carcasses are air chilled water is misted over them, while air blown over cooling elements is circulated throughout the room at a fairly high speed (Barbut, 2002). Advantages include drier carcasses and reduced exudation once packaged (Carroll and Alvarado, 2008). James et al. (2006) found carcasses lose an average of 1 - 1.5% total weight, with some as much as 3%. Research found less microbial contamination occurs between carcasses during air chilling compared to immersion (Barbut, 2002; Sanchez et al., 2002). This is possibly due to carcasses being hung individually on the processing line and unable to touch each other (Fluckey et al., 2003; James et al., 2006). Another possibility is the dehydration of the skin's surface caused by air chilling, injuring and killing bacteria (Berrang et al., 2008; Carroll and Alvarado, 2008). Rouger et al. (2017) suggested another reason for reduced microbial contamination being from low

refrigeration temperatures slowing down development of total viable bacteria, as well as inhibiting *Salmonella* and *Campylobacter*. The European Union does not allow carcasses to be sprayed during air chilling, as they feel it leads to aerosols of bacteria being spread by fans blowing chilled air throughout the room (Mead et al., 2000).

The European Union (EU) market introduced a new type of air chilling system, combining benefits of air and immersion chilling (Demirok et al., 2013). Carcasses remain on shackles and temperatures are reduced in countercurrent dip tanks followed by air chilling. Advantages to this system include reduced water usage of up to 95% and lowered energy costs up to 45% (Demirok et al., 2013). This system would be very beneficial to poultry processors, as immersion chilling is known for being very expensive and requiring large volumes of water. The combination system rapidly decreases carcass temperature and chilled products are shown to have increased shelf life and excellent quality (Demirok et al., 2013).

Pathogen Testing Strategies

Governmental agencies set requirements and standards to ensure products are as safe for consumers as possible. Strategies used to evaluate the presence of foodborne pathogens are essential to determining appropriate intervention methods. These methods and intervention requirements for each meat industry vary, as well as the protocols used in different parts of the world. There are very different testing strategies for the U.S. compared to those commonly used in Europe. Both strategies have advantages and disadvantages.

United States

The USDA-FSIS implements specific methods for processing plants to evaluate microorganism prevalence. The most common method used in the U.S. is whole carcass rinsing for evaluation of foodborne pathogens. Carcasses are required to be chosen for testing randomly and removed at specified points during processing procedures. Each carcass is placed in a bag with 400 mL sterile neutralizing buffered peptone water (BPW) and is hand shaken for 1 minute. At least 100 mL of this rinsate is collected into a sterile specimen jar and held at refrigeration temperatures until being shipped to a laboratory for analysis. Once rinse samples arrive at the laboratory, they are used for a variety of common foodborne pathogen testing, each requiring specific methods and confirmation techniques (Russell, 2012a).

Europe

The testing procedures used in Europe vary greatly from those in the U.S. While the U.S. evaluates whole carcasses, Europe pools neck skin samples from three individual birds (total 25 g) to be evaluated as a group (Cason et al., 2010; Russell, 2012a). All skins are placed into a stomacher bag together with 9X volume of a maximum recovery diluent. Each bag is homogenized for 1 minute and contents used for evaluation of different foodborne pathogens.

An alternate European sampling method for some countries utilizes a blowtorch or iodine solution for skin sterilization. Tissue samples are removed from deep within the muscle using sterile tweezers (Russell, 2012a). This method is often used to evaluate the presence of *Salmonella*, although positive samples are never found. Companies then

report themselves as “*Salmonella* free” to consumers. This tactic is extremely misleading though, as *Salmonella* is still likely present on carcass surfaces, but not found in deep tissue samples (Russell, 2012a).

United States Versus Europe

Processing techniques and microbial testing in the U.S. versus Europe differ in many aspects including the use of antimicrobials, chilling methods, etc. The EU has not permitted antimicrobial use in poultry since 1971 (Rio et al., 2007). Hygienic regulations in EU allow only potable water or steam for decontamination (Rio et al., 2007). The goal is not to allow processors to use antimicrobials to cover any unhygienic methods.

Procedures also differ for evaluating microbial loading on carcasses and parts within the processing facility. U.S. companies are required to post testing results which indicate which companies are below average and include information including prevalence, name, address, and USDA plant numbers online for the public (Russell, 2012b). This allows consumers full access to information regarding companies, to make educated purchasing decisions. It also gives companies the incentive to strive for lower pathogen prevalence within their facility, and to prevent negative publicity and potential customer loss.

Testing methods used by the U.S. versus Europe each have advantages and disadvantages. The U.S.’s whole carcass rinse method allows a carcass to be sampled entirely, both internally and externally, although it does not detect firmly attached *Salmonella*. Incubation steps are also required in detection for increased sensitivity and better opportunity to detect pathogen presence, even at low numbers (Russell, 2012b). A disadvantage to this method is each carcass having to be physically removed from the

processing line and either disposed of after being rinsed or undergoing reprocessing procedures. The European neck sampling method allows carcasses to remain on the processing line and continue processing procedures, as neck skin is clipped when birds pass sample collectors (Russell, 2012b). This method is physically easier and pooling neck skins more cost efficient, but only samples a small percentage of the carcass's surface.

Each method gives the possibility for false negatives, and Cox et al. (2010) demonstrated pathogens may be detected in one method but not the other and vice versa. The small amount of rinsate required in the U.S. method (only 7.5% in *Salmonella* testing) increases opportunities for pathogens to go undetected (Cox et al., 2010). Hutchinson et al. (2006) determined an advantage to European methods was no manual shaking required. It may be variable, and results possibly altered from different shaking methods or sampler fatigue. Researchers have shown the whole carcass method to be more sensitive and have the ability to find more positive samples compared to pooling neck skins (Hutchinson et al., 2006).

Marker Strain - Campylobacter

Antimicrobial resistant *Campylobacter* species have continued to emerge, and researchers have isolated them from food and water sources throughout the U.S., Europe, and Canada (Gaudreau and Gilbert, 1998; Smith et al., 1999; Talsma et al., 1999; Centers for Disease Control and Prevention, 2000; Saenz et al., 2000; Moore et al., 2001). Ge et al. (2003) found 94% of samples evaluated were contaminated with a *Campylobacter* strain resistant to at least one of the seven antibiotics used in the study. Research has

indicated strains resistant to antibiotics cause prolonged and more severe illnesses than strains susceptible to antibiotics (Travers and Barza, 2002; Moore et al., 2006). While *Campylobacter* infections are usually treated with fluid replenishment, antibiotics are sometimes used in patients with severe or prolonged infections (Blaser, 1997; Altekruuse et al., 1999). Therefore, *Campylobacter*'s growing susceptibility to antibiotic treatments makes it an even more serious concern.

Either chromosomes or plasmids usually mediate antibiotic resistance occurrence. Chromosomal resistance may evolve naturally or be due to stress on the organism (Cox et al., 2009a). When genes resistant to an antibiotic are generated, chromosomal loci or plasmid exchange occurs to transfer this new information to the bacterial isolates (Allen et al., 2007). Researchers have developed marker strains using these mechanisms to aid in the evaluation and detection of pathogens. These mutant strains can be used to inoculate samples and when once plated should be the only strain detected and able to grow on specified plating media made using the resistant antibiotic. This should allow for direct counting and eliminate the problem of background microflora present on samples prior to inoculation.

Cox et al. (2009a) developed a mutant marker strain of *Campylobacter coli* resistant to the antibiotic gentamicin to be used in poultry research. Gentamicin generally eliminates *Campylobacter* strains, therefore developing a mutant strain resistant to its effects makes an ideal marker organism (Ge et al., 2003; Gupta et al., 2004). Cox et al. (2009a) incorporated different levels of gentamicin into Campy-Cefex agar and first recovered the mutant strain in 2003. Since, it has been transferred numerous times and has maintained resistance to gentamicin. This resistance occurs from production of

aminoglycoside-modifying enzymes, and it is now a stable marker (Saenz et al., 2000; Lee et al., 2002; Shakil et al., 2008).

The Addition of Processing Aids

A variety of intervention strategies are used to reduce microbiological hazards during processing procedures (Stopforth et al., 2007). The most common method for finished carcasses is a multi-hurdle approach using multiple intervention strategies and aids (Leistner and Gorris, 1995; Bacon et al., 2000). The use of multiple intervention points throughout processing showed significant reductions in pathogens, allowing some facilities to meet performance standards required by FSIS (Stopforth et al., 2007).

Individual intervention points were also found to significantly reduce bacteria when used in processing water (Stopforth et al., 2007). Dips or sprays of chlorinated water, acidified sodium chlorite (ASC), cetylpyridinium chloride (CPC), chlorine dioxide, peroxyacetic acid (PAA), trisodium phosphate (TSP), acetic acid or lactic acid can be used to reduce *Campylobacter* levels on broiler carcasses (Stopforth et al., 2007; FAO, 2009). Studies demonstrated when dips or sprays are applied using such chemicals at different processing points, significant reductions can be made in microorganism prevalence (Stopforth et al., 2007). Processing facilities are required to conduct testing to validate intervention strategies used are effective in reducing pathogen presence (Stopforth et al., 2007).

Antimicrobials

Antimicrobials are an important addition to poultry processing and assist in reducing foodborne pathogens and cross contamination within facilities. A number of chemicals are approved for use in processing including acidified sodium chlorite (ASC), cetylpyridinium chloride (CPC), chlorine, chlorine dioxide, peroxyacetic acid (PAA), trisodium phosphate (TSP) or electrolyzed NaCl (Stopforth et al., 2007). Although many chemicals have been evaluated as a possible intervention to reduce pathogens, few are used commercially (Dickson and Anderson, 1992; Corry et al., 1995). To be used in the industry, antimicrobials must be approved, have documented efficacy, and appropriate concentrations and contact times for the processing location they are being considered (Bauermeister et al., 2008). An antimicrobial must also be cost efficient for the processing facility and not cause any form of organoleptic damage to the product or corrosion to machinery (Bauermeister et al., 2008). Antimicrobials have a minimum inhibitory concentration to determine bacteriostatic activity, meaning the minimum concentration of the chemical that completely inhibits growth of a targeted pathogen (Dubois-Brissonnet, 2012). Intervention strategies usually include antimicrobials being added to either a rinse or spray. Contact time and the concentration required are big factors to whether particular chemicals would be better suited as a rinse, dip, or spray (Bauermeister et al., 2008). If higher concentrations are required for adequate bacterial reductions, chemicals will need to be very cost effective or processors will choose an alternate method (Bauermeister et al., 2008). It is important to ensure an antimicrobial, when used at its proposed time, location, and concentration does not alter carcass quality in any way (Bauermeister et al., 2008).

Stopforth et al. (2007) determined no single intervention could significantly reduce the hazards of pathogens throughout the process. For adequate reductions on finished carcasses, a multi-hurdle approach must be implemented using several intervention methods throughout processing procedures (Bacon et al., 2000). Interventions often use organic acids including lactic acid and citric acid (SCVMRPH, 1998). Organic acids were shown to work well in eliminating bacteria, due to their ability to penetrate cell membranes and acidify cell contents, killing the bacteria (SCVMRPH, 1998). The variety of antimicrobials used throughout processing have different mechanisms of action and levels of efficacy. The effectiveness of chemicals may vary depending on microbial load and the composition of flora present (Gill and Badoui, 2004). Therefore, it should not be assumed every antimicrobial will have identical effects on meat from different sources. A tremendous amount of research has been done to evaluate these chemicals, the bacteria they affect and the advantages and disadvantages of their use.

Chlorine

Chlorine is a well-recognized treatment for poultry processing facilities and has been used at various points in the process for decades. It reduces bacteria, controls pathogen spreading, and prevents bacteria build-up on equipment and surfaces (Bailey et al., 1986). When added to water, chlorine binds to other chemicals, changing its chemical structure and making it less effective in reducing bacteria. Chlorine continues reacting with the impurities in water until water becomes oxidized, a process which affects the amount of free residual chlorine remaining to attack bacteria (Keener et al., 2004).

Remaining free chlorine changes to hypochlorous acid, the form of chlorine responsible for antimicrobial reductions (Gavin and Weddig, 1995). The reduced free residual chlorine leads to problem for chlorine added to the chiller, as chillers may contain a lot of organic material such as blood, digesta, and fat. Once chlorine binds to these materials, the availability of free chlorine remaining may be so small it is relatively useless as a chiller treatment.

Another disadvantage of using chlorine is its sensitivity to changes in water quality, pH, or addition of heat. When water pH rises above 7.0, chlorine quickly begins to lose effectiveness (SCVMRPH, 1998). Once pH of water reaches 8.0, chlorine is no longer found in its active hypochlorous acid form and is no longer effective for reducing bacteria, even at high levels (Hui and Sherkat, 2005). The pH of chiller water can rise from the carryover of high pH water remaining on birds leaving the scalding treatment (Berrang et al., 2011). Therefore, chlorine used in chiller water must be monitored and maintain a low pH or it will be ineffective (Berrang et al., 2011). One suggestion to address this problem is the addition of CO₂ gas aeration systems being added to the chiller, to reduce pH and allow chlorine to form hypochlorous acid. This may be too costly for some processing facilities. Another disadvantage of chlorine is the contact time it requires to act on bacteria, which may range from 5 - 15 minutes (Keener et al., 2004). This extended time requirement makes an unlikely quick treatment method for processing other than during chilling.

USDA-FSIS requires processors to use 20 - 50 ppm chlorine in the chiller to reduce cross contamination on carcasses, although research evaluating its effectiveness varies (Stopforth et al., 2007). Chlorine causes cell death by oxidizing components within

the cell (Oyarzabal, 2005). Some studies showed chlorinated treatments beneficial for carcass cleanliness, but not significant to reducing the presence of *Campylobacter* (Northcutt et al., 2005; Parveen et al., 2007). Contrasting results from Loretz et al. (2010) showed inoculated *Campylobacter* parts reduced 0.5 - 3.0 log cfu/mL when treated with chlorine. Other data also showed reductions in *Campylobacter* from each individual chlorinated water wash (FAO, 2009). Studies found when chlorine is maintained at a lower pH, it was capable of reducing *Salmonella* incidence on poultry skin (Lillard, 1980; Berrang et al., 2011). Kemp et al. (2001) reported *Salmonella* unresponsive to a variety of intervention methods used in processing facilities, including chlorine. Therefore, further research is needed to determine the true advantages of using chlorine throughout the processing facility, as current research exhibits conflicting results.

Chlorine Dioxide

Although it seems “chlorine” would be very similar to chlorine dioxide, the two are very different chemicals and have different structures and properties. The initial form of chlorine dioxide was a liquid registered by the Environmental Protection Agency (EPA) in 1967 for use as a disinfectant and sanitizer (Taylor and Wohlers, 2004). Chlorine dioxide is extremely effective in eliminating bacteria, viruses, spores, and other disease-causing organisms. Its mode of action is to oxidize cell membranes and constituents, breaking down cell walls when used at high concentrations (Oyarzabal, 2005). It functions independently of pH, therefore only requires a fraction of the dosage chlorine does (Lillard, 1979). When used in the chiller, concentrations are not to exceed residual levels greater than 3 ppm (Liem, 2002; Curtis and Butler, 2009). Being less

sensitive to pH changes and operating in a pH range of 5.0 - 10.0, chlorine dioxide is minimally affected by organic matter (USDA, 2002). The time required for it to be effective is much quicker compared to chlorine, especially when pH remains high.

Chlorine dioxide is primarily used in the chill tank and leaves no residue (USDA-FSIS, 2008). USDA-FSIS requires chlorine to be added to the chiller, but permits chlorine dioxide as an alternative method for cross-contamination reduction (Stopforth et al., 2007). When chlorine dioxide was used as a dip, it was found to reduce *Salmonella* by 15 - 20% (FAO, 2009). Stopforth et al. (2007) showed that combining chlorine and chlorine dioxide in the chiller was much more effective than either treatment alone. One potential disadvantage to using chlorine dioxide is the possibility brown coloration may develop on the wings, caused by blood oxidation (Brown, 2003).

Peracetic Acid

Peroxyacetic or peracetic acid (PAA) is an organic peroxide with a very low pH, formed by equal mixtures of acetic acid and hydrogen peroxide (Baldry and Fraser, 1988; Bauermeister et al., 2008; USDA-FSIS, 2016a). It is known for its strong, pungent odor and when in concentrated form being highly corrosive and unstable (USDA-FSIS, 2016a). On September 19, 2001 the FDA amended food additive regulations on PAA, regarding it as safe for use as an antimicrobial on poultry carcasses and parts (FDA, 2000). Other industries use PAA as an antimicrobial, cleaner and sanitizer, but it is used in the meat industry for reducing bacteria and food spoilage on carcasses and parts (USDA-FSIS, 2016a). The oxidizing ability of PAA disrupts the permeability of the cell membrane and alters protein synthesis to lead to cell death (Oyarzabal, 2005). One

advantage to its use is extended shelf life of products without causing organoleptic damage (Bauermeister et al., 2008). PAA is popular as an additive to chiller and in post-chill immersion tanks (Wideman et al., 2016).

Disadvantages to using PAA are related to safety concerns for employees and others, in areas where the product is being used. Concentrated forms of PAA are corrosive and can cause irreversible damage to the skin and eyes (USDA-FSIS, 2016a). Therefore, it must be used in diluted form not to exceed 2,000 ppm. Even in diluted form, PAA can form a vapor and irritate the eyes, nose, or throat of persons in the area (USDA-FSIS, 2016a). It is essential that proper ventilation is maintained, and the chemical is monitored for possible mixing with other chemicals within floor drains or for overspray from nozzles or spray cabinets. Preventing overexposure to PAA must include minimizing the vapor and ensuring employees are unable to come into contact with it in concentrated form.

Research evaluating the effectiveness of PAA has found concentrations as low as 0.0025% reduce *Salmonella* levels, although higher concentrations were required to reduce the incidence of *Campylobacter* (Bauermeister et al., 2008). A 1.5 log cfu/mL decrease in *Campylobacter* was shown when PAA was applied at 0.02% (Bauermeister et al., 2008). At higher concentrations, PAA applied in a post-chill dip tank was found to reduce *Salmonella* and *Campylobacter* by more than 2.0 log cfu/mL (Nagel et al., 2013). Further research indicated a 20 second post-chill dip caused no alterations to product quality, while significantly reducing pathogen presence (Nagel et al., 2013). When Chantarapanont et al. (2004) applied 0.01% solution of PAA on poultry skin for 15 minutes, only a 1.0 log reduction was found. Although most studies agree PAA

significantly reduces pathogens on poultry, different levels of effectiveness are reported, possibly attributed to a number of different factors between experiments such as contact time, initial microorganism level, etc.

Trisodium Phosphate

Trisodium phosphate (TSP) was a widely used antimicrobial in the past at various intervention points of poultry processing. The U.S. FDA determined TSP as “safe”, and the USDA approved its use as a food ingredient (Federal Register, 1982). The high alkalinity assists in removing fat films from the surface of meat and allows TSP to come into contact with more bacteria (Giese, 1993). The fat films are removed by disruption of the cell membrane, causing leaking of intracellular fluid (Capita et al., 2002). Once the cell begins leaking intracellular fluid, they are ultimately destroyed (Coppen, 1993; Giese, 1993). Once fat is removed, access to bacteria can also allow it to be more easily washed away from the surface (Bender and Brotsky, 1992; Giese, 1992; Kim et al., 1994; Keener et al., 2004). Korber et al. (1996) developed a way to measure plasmolysis within cells and show when a chemical damages membrane integrity to the point of cell death. This method was used to demonstrate how TSP damages the membrane by its high pH (Sampathkumar et al., 2004). TSP acts as a surfactant and the high pH allows its approval for use in online reprocessing (USDA-FSIS, 2008). One disadvantage is the concentration required (10%), thus making it a costly option for processors.

TSP has been shown effective in reducing pathogen levels at various points in poultry processing. A 15 second dip application at pH 12 reduced *Campylobacter* up to 1.7 log cfu/g (FAO, 2009). Kim et al. (1994) used a scanning electron microscope to

demonstrate that TSP removed *Salmonella* from chicken skin. An 8 - 12% solution used as a pre- or post-chill dip reduced *Salmonella* and *Campylobacter* 1 - 2 log cfu/mL (FAO, 2009). Results varied on studies evaluating TSP effectiveness in the chiller. If residual TSP remaining on carcasses enters the chiller, it dramatically increases pH levels beyond the standard range of 9.7 - 10.5. This prevents chlorine in the chiller from being converted to its active form, hypochlorous acid. Chlorine is effective at pH 6.5 - 7.5, therefore chlorine would be ineffective and processing plants would be wasting the chlorine being added to the chiller, essentially an expensive problem. A suggestion to counteract this pH increase is to add phosphoric acid to the chiller, although cost would greatly increase (Arritt, 2000). Some plants using TSP dip systems experienced increased *Salmonella* levels, possibly from TSP washing *Salmonella* from one carcass onto others. Although it may reduce *Salmonella* numbers on single carcasses, overall prevalence may increase and force corrective actions from the USDA.

Scientists questioned if high pH was the main factor for TSP's success. When pH was adjusted to 7.0, no pathogen reductions were shown, therefore the alkaline pH was found directly correlated to its effectiveness (Sampathkumar et al., 2004). Although TSP at high pH is effective, it can cause problems in the processing facility including corrosion to equipment or increased phosphoric acid in chiller water causing potential problems at water treatment plants (Arritt, 2000; Brown, 2003). TSP also has a level 3 hazard rating, meaning it is capable of causing severe or permanent damage to eyes, skin and lungs of plant employees (Arritt, 2000). Therefore, in spite of TSP effectiveness in reducing foodborne pathogens, expenses and further issues within the processing facility are possibilities.

Acidified Sodium Chlorite

The U.S. FDA approved the use of acidified sodium chlorite (ASC) as an antimicrobial treatment on poultry, red meat, seafood, fruits and vegetables. ASC is produced by a solution of sodium chlorite and a GRAS (generally recognized as safe) acid. Citric acid is commonly used and shown more effective than phosphoric acid in eliminating microorganisms, such as *Campylobacter* (Kemp et al., 2000). Once a solution is mixed, and pH drops below 4.0, chlorite dissociates to chlorous acid (Keener et al., 2004). ASC acts as a disinfectant and oxidizes cell walls by attacking sulfide and disulfide protein linkages (Keener et al., 2004; Oyarzabal, 2005). When being mixed, the goal is to achieve a pH of 2.3 - 2.9 for successful reprocessing of products (Kemp et al., 2001).

One constraint of ASC is its limited ability to penetrate beneath the surface of a carcass, allowing embedded organisms to remain untouched (Kemp et al., 2001). ASC was originally applied pre-chill in a vented cabinet at concentrations up to 1,200 ppm (Brown, 2003). Stopforth et al. (2007) found dipping poultry products in 500 - 1,200 ppm ASC significantly reduced overall bacterial loads and *Salmonella*. Other research indicated ASC should be used at 600 - 800 ppm (pH 2.5 - 2.7) for immersing whole carcasses for 15 seconds to reduce bacteria on the skin's surface (FAO, 2009). Prior to chilling, both a 15 second spray and 5 - 8 second dip of ASC were shown to significantly reduce *Salmonella* or *Campylobacter* levels and allowed even greater reductions when followed by a freshwater wash (FAO, 2009). When used specifically in chiller water, ASC is limited to 50 - 150 ppm and can be used with other GRAS chemicals.

Campylobacter was lowered by 2.6 log cfu/mL when whole carcasses were immersed post-chill (FAO, 2009). Some processors switched from TSP to ASC for reprocessing, as it was shown to be more effective.

Cetylpyridinium Chloride

Cetylpyridinium chloride (CPC), also referred to as Cecure, is a stable, quaternary ammonium compound that is non-volatile, soluble in water, and approved for use in ready to cook products (USDA-FSIS, 2008). It was previously used as an ingredient in mouthwashes, toothpastes and throat lozenges (Smith et al., 1991; Beers et al., 2006). The FDA approved it to be used as an antimicrobial processing aid on poultry carcasses pre-chill in 2004 (Beers et al., 2006; Gilbert et al., 2015). The FDA granted an amendment in 2007, allowing raw carcasses to be treated post-chill with CPC (Gilbert et al., 2015). Wideman et al. (2016) demonstrated the ability of this treatment to reduce *Campylobacter* to undetectable levels. Beers et al. (2006) demonstrated spraying carcasses pre-chill with CPC significantly reduced the presence of *Salmonella*, *Campylobacter*, APC counts, *E. coli*, and coliforms. Further research demonstrated its ability to reduce *C. jejuni* on poultry skin by greater than 4.2 log cfu/mL (Riedel et al., 2009). These reductions may be due to the inactivation of the compound in the rinsate or residual rinse fluid. Many researchers showed Cecure to be significantly more effective against *Campylobacter* at concentrations of 0.25 - 0.5% (Li et al., 1997; Waldroup et al., 2000; Arritt et al., 2002; Oyarzabal, 2005; Beers et al., 2006).

The pH of CPC is relatively neutral and research has shown it does not cause organoleptic damage to the product (USDA-FSIS, 2008). The hydrophilic portion reacts

with cell membranes, causing cellular components to leak from the cell (Oyarzabal, 2005). This disrupts cellular metabolism and can result in death of the cell. An advantage to CPC's use is it was not found to be a severe health hazard, corrode metal, or cause phosphate waste as previously shown problems with the use of TSP (Arritt, 2000). Bai et al. (2007) showed a 1-day extension of shelf life when spraying with 0.4 - 0.5% (4000 – 5000 ppm) Cecure, extended to 2 days using 1.0 - 1.5% (10,000 – 10,500 ppm) concentrations on boneless, skinless thighs. Researchers showed concern as to whether these extensions were a result of initial treatment or continued action of the chemical during refrigerated storage (Bai et al., 2007). This was evaluated by Gilbert et al. (2015), finding a slight reduction on the initial day of treatment and determining Cecure does not result in continued chemical effect on aerobic plate counts during storage.

Organic Acids

Organic acids have been evaluated for use as antimicrobials throughout poultry processing procedures (Izat et al., 1990; Tamblyn and Conner, 1997; Bilgili et al., 1998). Some organic acids commonly investigated are acetic, formic, citric, lactic and propionic (Mulder et al., 1987; Izat et al., 1990; Dickens et al., 1994). A disadvantage to using organic acids is their decreased effectiveness on bacteria firmly attached to broiler skin (Tamblyn and Conner, 1997). One huge disadvantage seems to be the visual quality changes within meat after being treated with organic acids (Mulder et al., 1987; Izat et al., 1990; Dickens and Whittemore, 1995). Dickson and Anderson (1992) related these changes to concentration, contact time and temperature when treated. Bilgili et al. (1998) showed using different organic acids, even at reduced concentrations, altered colorimeter

values by decreased lightness and increased redness and yellowness colorations of broiler skin. Other researchers demonstrated similar results, also showing altered visual appearance in broiler carcasses treated with organic acids (Mulder et al., 1987; Izat et al., 1990; Dickens et al., 1994). Increased concentrations were correlated to decreased redness colorimeter values (Bilgili et al., 1998). Dickens et al. (1994) reported darkening and yellowing of carcasses when most organic acids were used.

Meat color is an important factor in consumer purchasing decisions; therefore any alterations are likely to push processors toward other antimicrobials (Djenane et al., 2003). Other visual defects including surface bleaching and brown discoloration. Bautista et al. (1997) attributed such coloration to oxidation reactions. In addition, off-odors were shown to occur from organic acid use at concentrations greater than 1.5 - 2.0% (Smulders, 1995; Ellerbroek et al., 1996; Zeitoun et al., 1996; Tamblyn and Conner, 1997; Huffman, 2002; Mehayar et al., 2005).

Although organic acids have been shown to be successful in reducing microbial contamination, combining them with antimicrobials may assist in avoiding quality changes caused by the acid use alone (Bauermeister et al., 2008). When combining chemicals, lower concentrations of organic acids can be used and visual alterations reduced, while still retaining effective antimicrobial capabilities (Bauermeister et al., 2008). Brinez et al. (2006) found the combination of organic acids and hydrogen peroxide effective. This was supported by previous research showing 3 log cfu/mL reductions on carcasses sprayed with a solution of 1% acetic acid and 3% hydrogen peroxide (Bell et al., 1997).

Citric Acid

Citric acid has been evaluated for use in poultry processing facilities and diffuses through cell membranes to penetrate weak non-dissociated acids. This acidifies the cytoplasm of the cell, as the acid accumulates, and disrupts the proton motive force making the cell unable to transport substrates (Vasseur et al., 1999). Vasseur et al. (1999) demonstrated the highest inhibitory effect due to citric acid's ability to diffuse through cell membranes. Wideman et al. (2016) found citric acid use in the scalding not effective and attributed this to the high levels of organic matter within the scalding, altering the acids effectiveness. Citric acid has been shown to reduce pathogens 1.9 log cfu/mL when used in a chiller tank at 4% compared to control samples (Ricke et al., 2012). Another study using the same conditions only showed a 0.7 log cfu/mL reduction (Tamblyn and Conner, 1997). Since organic acids are known to cause organoleptic damage to poultry, Gonzalez-Fandos et al. (2009) reported a 3% concentration produced no unacceptable odors or color changes.

Lactic Acid

Lactic acid was shown to penetrate the cytoplasmic membrane and alter pH within the cell and disrupt proton motive force (Alakomi et al., 2000). This force is disrupted when acid decreases the ionic concentration of the cell membrane, causing acid accumulation in the cell (Vasseur et al., 1999). Once the force is disrupted, substrate transport is ceased, and the cell is destroyed. As organic acids are known to cause color changes in meat products, Deumier (2004) evaluated poultry skin treated with lactic acid and observed a yellow/greenish appearance. In contrast, many studies found using 1 - 2%

lactic acid is effective in reducing bacterial presence, without altering any organoleptic characteristics of the meat (Anderson and Marshall, 1990; Smulders, 1995; USDA-FSIS, 1996; Dincer and Unluturk, 2001).

Research evaluating the efficacy of lactic acid to reduce *Campylobacter* shows inconsistent results (Zhao and Doyle, 2006; Lecompte et al., 2009; Riedel et al., 2009; Rajkovic et al., 2010). Rasschaert et al. (2013) studied two methods of application, spraying versus dipping, and found pathogen reduction significant when carcasses were submerged (1.24 - 1.62 log cfu/mL). This reduction was attributed to spray treatments not allowing a sufficient amount of acid to cover the carcass. Findings have indicated a wide range of reductions of 0 - 4 log cfu/mL (Lecompte et al., 2009; Riedel et al., 2009; Rajkovic et al., 2010). Differences between studies may be concentrations, exposure times, contamination levels, or application techniques.

Campylobacter

Campylobacter is considered one of the most common causes of bacterial gastroenteritis in the world, commonly transmitted through poultry and poultry products (Atabay and Corry, 1997; Blaser, 1997; Allos, 2001; Kessel et al., 2001; Cox et al., 2010). Studies found 50 - 70% of *Campylobacter* cases to be associated with raw poultry and are usually sporadic in occurrence (Tauxe, 1992; Allos, 2001; Humphrey et al. 2007). Researchers found high levels of *C. jejuni* present in retail chicken, indicating a source for potential infections (Harrison et al., 2001; Moore et al., 2002). Symptoms of campylobacteriosis are fever, abdominal pain and diarrhea, typically occurring within 2 - 5 days of ingesting contaminated food (Robinson, 1981; Black et al., 1988). Treatment

generally requires high intake of fluids and symptoms usually subside in 3 - 10 days, although severe cases may require antibiotics (Keener et al., 2004). Fatalities are not common from *Campylobacter*, although people do die from secondary complications, such as septicemia, meningitis, or appendicitis (Blaser, 1997; Saleha et al., 1998).

Campylobacter has a very low infectious dose; as few as 500 organisms can lead to illness (Robinson, 1981; Black et al., 1988; Friedman et al., 2000). Only one drop of raw chicken juice is needed to cause campylobacteriosis in an individual (Friedman et al., 2000; Newell and Wagenaar, 2000). *Campylobacter* is more common in children less than a year old, young adults from 15 - 25 years old and individuals with suppressed immune systems (Friedman et al., 2000; Keener et al., 2004).

Campylobacter are gram-negative bacteria, appearing as slender, spiral curved rods under the microscope often described as a “comma”. They typically range from 0.2 - 0.8 μm wide and 0.5 - 5.0 μm long (Keener et al., 2004). *Campylobacter* are known for their distinctive rapid movements, described as darting and spinning in place. Movement is accomplished from one flagellum at each end, allowing them to travel to attachment sites and penetrate intestinal cells (Cox et al., 2010). If unable to move, the organism no longer has the ability to colonize the gastrointestinal tract or cause illness (Cox et al., 2010). Growth is under thermophilic conditions, optimally at temperature ranges of 37 - 42°C, but incapable of growth under 30°C (Keener et al., 2004). When grown on plated media, some colonies appear smooth, convex and glisten. Others are flat, translucent and irregularly shaped and all are usually colorless, gray, or light cream colored (Cox et al., 2010). The specific species found to most commonly cause gastroenteritis in humans is

C. jejuni, while *C. coli* is also responsible for much of the illnesses (Nachamkin, 1995; Cox et al., 2010; USDA-FSIS, 2014).

Campylobacter is commonly found in the intestinal tract of mammals and birds and shed through feces (FAO, 2003; FAO, 2009). This allows it to be spread across various parts of the environment, although it must eventually find a host to inhabit (FAO, 2009). Being a commensal organism, it often remains undetected as birds show no visible symptoms once infected. When introduced in a poultry flock, it spreads very quickly and remains colonized within birds until processing (Jacobs-Reitsma et al., 1995; Gregory et al., 1997). *Campylobacter* has become a very serious concern throughout the poultry industry with prevalence as high as 70% in processing facilities and the majority of flocks found to be *Campylobacter* positive (Zhao et al., 2001; Williams and Oyarzabal, 2012). In 2012, a study found 21% of poultry parts were contaminated with *Campylobacter* (USDA-FSIS, 2015), while NARMS reported 38% of retail chicken was positive in 2013 (NARMS, 2015).

Dangers and Spreading

In 1968, *Campylobacter* was first isolated from the stool samples of patients with diarrhea (Butzler, 2004). *Campylobacter* was identified as a human diarrheal pathogen in 1973 (Altekruse et al., 1999). It became known as one of the leading causes of bacterial gastroenteritis in the world by the 1980's (Allos, 2001). Currently, *Campylobacter* is estimated to cause 850,000 illnesses annually in the U.S. (USDA, 2017b). This leads on average to more than 8,000 hospitalizations and 80 deaths per year (USDA, 2017b). One reason *Campylobacter* can be so dangerous is the risk of developing Guillain-Barre

syndrome after being infected, although this is rare (Nachamkin et al., 1998; McCarthy and Giesecke, 2001). This syndrome is an autoimmune disorder leading to temporary paralysis with the possibility of permanent nerve damage. The low infectious dose of *Campylobacter* is very problematic and increases the likelihood for cross-contamination to occur during processing. Therefore, the industry began implementing antimicrobial additives to reduce chances of the pathogen spreading (Behravesch et al., 2012).

Campylobacter species with the ability to cause human illness continue to emerge (Butzler, 2004). These new species can be difficult to culture and often have complex nutrient requirements (Lastovica, 2006), allowing them to remain undetected in some cases. Undiscovered species may be responsible for a large number of gastrointestinal illnesses, making *Campylobacter* an even greater concern than it is currently (Man, 2011).

The antimicrobial resistance of *Campylobacter* species is another concern for consumers and processors. In 1995, the FDA approved fluoroquinolones to be used in poultry and quickly after, scientists discovered a strain of *Campylobacter jejuni* resistant to quinolone (Engberg et al., 2001). The quinolone resistant strain was found to cause 1.3% of *Campylobacter* illnesses in 1992, 10.2% by 1998 and was located in 14% of the retail chicken sampled by 1997 (Smith et al., 1999). Evaluating this issue internationally, showed farms using little to no fluoroquinolone had few resistant infections present, while farms using large amounts of fluoroquinolones were more likely to isolate the resistant strain from human and animal samples (Piddock, 1999; Nelson et al., 2007). These findings influenced the FDA's decision to withdraw the use of fluoroquinolones for poultry drinking water in 2005 (NARMS, 2015).

Survival Techniques

There are some environmental challenges *Campylobacter* must overcome prior to entering a host, such as exposure to oxygen, low temperatures, desiccation and other stress factors (Park, 2002). Although *Campylobacter* is unable to grow or reproduce when not inhabiting a host, it has the unique ability to survive (Murphy et al., 2006), making it an even more dangerous pathogen for humans. *Campylobacter* cells can enter a viable, but nonculturable state when presented with unfavorable conditions (Rollins and Colwell, 1986). As cells age, they become coccoidal shaped and unculturable (Moran and Upton, 1986; Rollins and Colwell, 1986; Jones et al., 1991; Pearson et al., 1993). Pearson et al. (1993) used direct immunofluorescence microscopy to observe these changes in different water supplies. Once broilers ingested contaminated water, *Campylobacter* returned to its spiral form with the ability to inhabit and colonize the intestines of the bird (Rollins and Colwell, 1986). Therefore, cells may be metabolically active with respiratory activity, but remain unable to be cultured and then recover only when reaching a susceptible host (Cox et al., 2010). Research determined another reason *Campylobacter* is successful is its demonstration of gene expression responses when presented with environmental stress (Park, 2002). These responses were very limited in comparison to other foodborne bacteria. This extraordinary survival technique makes *Campylobacter* one of the most dangerous pathogens and able to survive in a wide range of environments (Park, 2002). Murphy et al. (2006) attributes its survival as the greatest causative agent of bacterial foodborne illness to humans.

Poultry remains the most common, viable host for *Campylobacter*, as the high body temperature is an ideal environment for the pathogen (Skirrow, 1977). Studies have isolated *Campylobacter* from many water sources including rivers, estuaries, and coastal waters, ranging in populations of 10 - 230 cfu/mL (Bolton et al., 1982; Bolton et al., 1987). Buswell et al. (1998) found it able to survive in groundwater for several weeks. *Campylobacter* must be carried through the environment, via insect or vermin, although humans remain the most common route of transportation (Ridley et al., 2008). Cox et al. (2010) determined the bacteria could survive at -20°C on chicken carcasses for more than 64 days and on livers more than 84 days. Interestingly, *Campylobacter* spp. are eliminated more quickly at 25°C than either 4°C or 30°C (Cox et al., 2010). *Campylobacter* have a genome variation giving genome plasticity, assisting their adaptation and survival in hostile environments. If extracellular proteins are secreted during growth, they are shown to have higher stress tolerances (Murphy et al., 2003). These are some reasons *Campylobacter* is such a fascinating pathogen, with the unique ability to keep itself alive until infecting and thriving within an environmentally stable host.

Colonization Within the Bird

Researchers have evaluated the movement and colonization of *Campylobacter* after it enters a broiler. It is known to colonize the intestinal tract, specifically the ceca, although also found in other organs and tissues (Cox et al., 2006; Keener et al., 2004). Research demonstrated *C. jejuni* and *C. coli* were recovered from 20% of spleen, 17% of liver-gallbladder and up to 32% of unabsorbed yolk sac samples (Cox et al., 2006; Cox et

al., 2007). Meinersmann et al. (1991) determined the main colonization point was the mucus layers in the lower intestinal tract that overlay the epithelial cells within the ceca and cloacal crypts. Another study found *Campylobacter* present in the circulating blood of broilers, indicating the possible path *Campylobacter* may use to disseminate to organs and other tissues (Richardson et al., 2011). If *Campylobacter* is circulating within the blood, it is likely the reason for cross contamination during processing procedures (Richardson et al., 2011).

Scientists have also looked at *Campylobacter* presence in broiler breeders, recovering it from numerous internal organs and tissues (Carmarda et al., 2000; Buhr et al., 2002). *Campylobacter* was prevalent in 26% of thymus, 19% of spleen and 9% of liver-gallbladders (Cox et al., 2006). Many researchers found *C. jejuni* and *C. coli* within the mature and immature ovarian follicles and all segments of the reproductive tract (Jacobs-Reitsma, 1997; Carmarda et al., 2000; Buhr et al., 2002; Cox et al., 2005a). Cox et al. (2005b) determined *Campylobacter* spp. present in 26% of mature and 12% of immature ovarian follicles. Buhr et al. (2002) demonstrated the number of positive samples increased when moving down the different segments of the reproductive tract toward the cloaca. These results were attributed to possible fecal retrograde contamination. When evaluating broiler breeder roosters, Cox et al. (2002) found over 9% of semen samples to be naturally contaminated with *Campylobacter* spp. Therefore, research indicated it is not limited to the intestinal tract and can be recovered throughout the bird including the lymphoid organs, ovarian follicles, liver and gallbladder, reproductive tracts and semen of broiler breeders (Cox et al., 2002; Cox et al., 2005b; Cox et al., 2006). It is also noted that although there are considerable differences in

management and husbandry practices between Leghorn laying hens and broiler breeders, isolation rates of *Campylobacter* were very similar between the two (Cox et al., 2009b).

Some scientists wanted to determine if birds were colonized with *Campylobacter* prior to leaving the hatchery as day old chicks. First, Clarke and Bueschkens (1985) inoculated fertile eggs with *Campylobacter* using pressure differential and showed 11% of chicks hatched had *Campylobacter* within their intestinal tract. Another study used colony DNA hybridization to find 35% of newly hatched chicks had *Campylobacter* within their cecal contents, therefore were contaminated before leaving the hatchery facility (Chuma et al., 1994). To evaluate how rapidly *Campylobacter* spreads through a chick, Cox et al. (2005b) inoculated chicks orally and intracloacally, finding *Campylobacter jejuni* spread to primary and secondary lymphoid organs within one hour and persisted for 7 days.

Spread Through Production

Campylobacter should be addressed prior to the arrival of birds to the processing facility and began preventative measures in the production phase. Researchers have found addressing pathogens while birds are still at the hatchery facility may reduce their prevalence throughout the production process. Russell (2012c) suggested treating hatching cabinets with disinfectant fogging systems, electrostatic spraying, or by spraying disinfectant every 30 minutes. Ricke (2014) proposed chemical treatments for hatching cabinets such as hydrogen peroxide, chlorine dioxide, or trisodium phosphate.

Many researchers have suggested strict hygienic measures for individuals entering a broiler production house to reduce the risk of *Campylobacter* contamination through a

flock (van de Giessen et al., 1996; Evans and Sayer, 2000; Hald et al., 2000; Gibbens et al., 2001). Farm workers are potential transmitters of *Campylobacter* when entering multiple houses without using preventative measures, while other vectors include rodents, flies or wild birds (Annan-Prah and Janc, 1988; Berndtson et al., 1996; Gregory et al., 1997; Hielt et al., 2002). Other treatments include additives for water supplies such as acetic, lactic, formic or other organic acids (Immerseel et al., 2009). Scientists have also suggested biological, physical or a combination treatment for poultry litter pathogen reduction (Zamora-Sanabria and Alvarado, 2017). Many intervention strategies exist for reducing *Campylobacter* prevalence during the hatchery and grow-out processes, although a strict biosecurity plan is essential to properly address such issues.

Seasonal Variation

Scientists found *Campylobacter* incidence often changes seasonally, being more prevalent in warmer months (Harris et al., 1986; Annan-Prah and Janc, 1988; Berndtson and Engvall, 1989; Hinton et al., 2004b). Over a 12-month period, contamination exhibited a cyclical pattern (Willis and Murray, 1997; Hudson et al., 1999). Suggested reasons were the increased ventilation and higher insect population during warmer periods (FAO, 2009). Increased ventilation could lead to more flies inhabiting broiler houses spreading *Campylobacter* through flocks (FAO, 2009). Although many studies worldwide demonstrated data supporting seasonal changes in *Campylobacter* (Kapperud et al., 1993; Jacobs-Reitsma et al., 1994; Newell and Davidson, 2003; Rosenquist et al., 2009), others found no differences (Gregory et al., 1997; Nadeau et al., 2002). Brennhovd et al. (1992) of Norway suggested *Campylobacter* spp. use water as a vehicle of

transmission, once finding its presence on surface water sources. It was further reported the presence of *Campylobacter* in water was greater during autumn, also supporting the idea of seasonality (Brennhovd et al., 1992). This contaminated drinking water was found to be one reason chicks showed higher levels of *Campylobacter* contamination (Kapperud et al., 1993).

Poultry Testing Procedures

USDA-FSIS provides detailed instructions on the procedures to evaluate and isolate *Campylobacter jejuni/coli/lari* in poultry from rinse, sponge, or raw product samples (USDA-FSIS, 2016b). Described in the methods are steps for qualitative direct plating and enrichment for isolating and identifying from sponge samples. Presumptive positive tests are examined under the microscope for typical morphology and motility. If a sample still appears positive, confirmation tests to confirm to the genus level are completed including catalase, oxidase and latex agglutination. Latex agglutination assays are commercially available and other automated kits and systems have been developed, some combining various identification approaches to evaluate *Campylobacter* (Cloak et al., 2001; Padungtod et al., 2002). If confirming species, samples must undergo biochemical and physiological testing, which are time consuming to complete. Typical biochemical characteristics are negative reactions to methyl red, acetone, and indole production and a positive reaction to oxidase activity (Vandamme, 2000; Sellars et al., 2002).

References

- Acuff, G. R., C. Vanderzant, M. O. Hanna, J. G. Ehlers, F. A. Golan, and F. A. Gardner. 1986. Prevalence of *Campylobacter jejuni* in turkey carcass processing and further processing of turkey products. *J. Food Prot.* 49(9):712-717.
- Alakomi, H. L., E. Skytta, M. Saarela, T. Mattila-Sandholm, K. Latva-Kala, and I. M. Helander. 2000. Lactic acid permeabilizes gram-negative bacteria by disrupting the outer membrane. *Appl. Environ. Microbiol.* 66(5):2001-2005.
- Allen, V. M., S. A. Bull, J. E. Corry, G. Domingue, F. Jorgensen, J. A. Frost, R. Whyte, A. Gonzalez, N. Elviss, and T. J. Humphrey. 2007. *Campylobacter* spp. contamination of chicken carcasses during processing in relation to flock colonization. *Int. J. Food Microbiol.* 113(1):54-61.
- Allen, V. M., D. B. Tinker, M. H. Hinton, and C. M. Wathes. 2003. Dispersal of microorganisms in commercial defeathering systems. *Br. Poult. Sci.* 44(1):53-59.
- Allos, B. M. 2001. *Campylobacter jejuni* infections: update on emerging issues and trends. *Clin. Infect. Dis.* 32(8):1201-1206.
- Altekruse, S. F., N. J. Stern, P. I. Fields, and D. L. Swerdlow. 1999. *Campylobacter jejuni* – an emerging foodborne pathogens. *Emerg. Infect. Dis.* 5(1):28-35.
- Anderson, M. E., and R. T. Marshall. 1990. Reducing microbial populations on beef tissues: concentration and temperature of an acid mixture. *J. Food Sci.* 55(4):903.
- Annan-Prah, A., and M. Janc. 1988. The mode of spread of *Campylobacter jejuni/coli* to broiler flocks. *J. Vet. Med.* 35:11-18.
- Anthony, N. B. 1998. A review of genetic practices in poultry: efforts to improve meat quality. *J. Muscle Foods.* 9(1):25-33.

- Arnold, J. W. 2007. Bacterial contamination on rubber picker fingers before, during, and after processing. *Poult. Sci.* 86(12):2671-2675.
- Arnold, J. W., and S. Silvers. 2000. Comparison of poultry processing equipment surfaces for susceptibility to bacterial attachment and biofilm formation. *Poult. Sci.* 79(8):1215-1221.
- Arnold, J. W., and I. E. Yates. 2009. Interventions for control of *Salmonella*: clearance of microbial growth from rubber picker fingers. *Poult. Sci.* 88(6):1292-1298.
- Arritt, F. M. 2000. Efficacy of selected chemicals on the attachment and survival of *Campylobacter jejuni* on chicken breast skin. Master's Thesis. Food Science Technology. Virginia Polytechnic Institute and State University.
- Arritt, F. M., J. D. Eifert, M. D. Pierson, and S. S. Sumner. 2002. Efficacy of antimicrobials against *Campylobacter jejuni* on chicken breast skin. *J. Appl. Poult. Res.* 11(4):358-366.
- Atabay, H. I., and J. E. L. Corry. 1997. The prevalence of campylobacters and arcobacters in broiler chickens. *J. Appl. Microbiol.* 83(5):619-626.
- Bacon, R. T., K. E. Belk, J. N. Sofos, R. P. Clayton, J. O. Reagan, and G. C. Smith. 2000. Microbial populations on animal hides and beef carcasses at different stages of slaughter in plants employing multiple-sequential interventions for decontamination. *J. Food Prot.* 63(8):1080-1086.
- Bai, Y., K. R. Coleman, C. W. Coleman, and A. L. Waldroup. 2007. Effect of cetylpyridinium chloride (Cecure CPC Antimicrobial) on the refrigerated shelf life of fresh boneless, skinless broiler thigh meat. *Int. J. Poult. Sci.* 6(4):91-94.
- Bailey, J. S., J. E. Thomson, and N. A. Cox. 1987. Contamination of poultry during

- processing. Pages 193-211 in the *Microbiology of Poultry Meat Products*. F. E. Cunningham and N. A. Cox, eds. Academic Press, Orlando, FL.
- Bailey, J. S., J. E. Thomson, N. A. Cox, and A. D. Shackelford. 1986. Chlorine spray washing to reduce bacterial contamination of poultry processing equipment. *Poult. Sci.* 65(6):1120-1123.
- Baker, R. C., and C. A. Bruce. 1995. Further processing of poultry. In *Processing of Poultry*. G. C. Mead, ed. Springer. Boston, M.A.
- Baldry, M. G. C., and J. A. L. Fraser. 1988. Disinfection with peroxygens. In *Industrial Biocides: reports on applied chemistry*. K. R., Payne, ed. John Wiley and Sons. Hoboken, N.J.
- Barbut, S. 2002. *Poultry products processing*. CRC Press. Boca Raton, FL.
- Batz, M. B., S. Hoffmann, and J. G. Morris. 2012. Ranking the disease burden of 14 pathogens in food sources in the United States using attribution data from outbreak investigations and expert elicitation. *J. Food Prot.* 75(7):1278-1291.
- Bauermeister, L. J., J. W. Bowers, J. C. Townsend, and S. R. McKee. 2008. The microbial and quality properties of poultry carcasses treated with peracetic acid as an antimicrobial treatment. *Poult. Sci.* 87(11):2390-2398.
- Bautista, D. A., N. Sylvester, S. Barbut, and M. W. Griffiths. 1997. The decontamination efficacy of antimicrobial rinses on turkey carcasses using response surface designs. *Int. J. Food Microbiol.* 34(3):279-292.
- Beers K., J. Rheingans, K. Chinault, P. Cook, B. Smith, and A. Waldroup. 2006. Microbial efficacy of commercial application of Cecure CPC antimicrobial to ingesta-contaminated pre-chill broiler carcasses. *Int. J. Poult. Sci.* 5(8):698-703.

- Behravesh, C. B., I. T. Williams, and R. V. Tauxe. 2012. Emerging foodborne pathogens and problems: expanding prevention efforts before slaughter or harvest. Institute of Medicine. Improving food safety through a one-health approach: workshop summary. National Academics Press (US). Washington, D.C.
- Bell, K. Y., C. N. Cutter, and S. S. Sumner. 1997. Reduction of foodborne microorganisms on beef carcass tissue using acetic acid, sodium bicarbonate, and hydrogen peroxide spray washes. *Food Microbiol.* 14(5):439-448.
- Bender, F. G., and E. Brotsky. 1992. Process for treating poultry carcasses to control *Salmonella* growth. U.S. Patent 5,143,739.
- Berndtson, E., M. L. Danielsson-Tham, and A. Engvall. 1996. *Campylobacter* incidence on a chicken farm and the spread of *Campylobacter* during the slaughter process. *Int. J. Food Microbiol.* 32:35-47.
- Berndtson, E., and A. Engvall. 1989. Epidemiological studies of factors affecting the occurrence of campylobacters in broiler farms. Pages 61-62 in the Proceedings of the fifth international workshop on *Campylobacter* infections. G. M. Ruiz-Palacios, E. Calva, B. R. Ruiz-Palacios, eds. Puerto Vallarta, Mexico.
- Berrang, M. E., R. J. Buhr, J. A. Cason, and J. A. Dickens. 2001. Broiler carcass contamination with *Campylobacter* from feces during defeathering. *J. Food Prot.* 64(12):2063-2066.
- Berrang, M. E., N. A. Cox, G. R. Gamble, R. J. Meinersmann, B. B. Oakley, and A. Hinton, Jr. 2018. Contributors to the 100-fold increase in *Campylobacter* numbers on broiler carcasses during defeathering. *Poultry USA*. Watt Publishing Company. In Press.

- Berrang, M. E., and J. A. Dickens. 2000. Presence and level of *Campylobacter* spp. on broiler carcasses throughout the processing plant. *J. Appl. Poult. Res.* 9(1):43-47.
- Berrang, M. E., R. J. Meinersmann, D. P. Smith, and H. Zhuang. 2008. The effect of chilling in cold air or ice water on the microbiological quality of broiler carcasses and the population of *Campylobacter*. *Poult. Sci.* 87(5):992-998.
- Berrang, M. E., W. R. Windham, and R. J. Meinersmann. 2011. *Campylobacter*, *Salmonella*, and *Escherichia coli* on broiler carcasses subjected to a high pH scald and low pH postpick chlorine dip. *Poult. Sci.* 90(4):896-900.
- Bilgili, S. F., D. E. Conner, J. L. Pinion, and K. C. Yamblyn. 1998. Broiler skin color as affected by organic acids: influence of concentration and method of application. *Poult. Sci.* 77(5):752-757.
- Bilgili, S. F., A. L. Waldroup, D. Zelenka, and J. E. Marion. 2002. Visible ingesta on pre-chill carcasses does not affect the microbiological quality of broiler carcasses after immersion chilling. *J. Appl. Poult. Res.* 11(3):233-238.
- Black, R. E., M. M. Levine, M. L. Clements, T. P. Hughs, and M. J. Blaser. 1988. Experimental *Campylobacter jejuni* infection in humans. *J. Infect. Dis.* 157(3):472-479.
- Blaser, M. J. 1997. Epidemiologic and clinical features of *Campylobacter jejuni* infections. *J. Infect. Dis.* 176(Suppl. 2):S103-S105.
- Bolton, F. J., D. Coates, D. N. Hutchinson, and A. F. Godfree. 1987. A study of thermophilic campylobacters in a river system. *J. Appl. Bacteriol.* 62(2):167-176.
- Bolton, F. J., P. M. Hinchliffe, D. Coates, and L. Robertson. 1982. A most probable

- number method for estimating small numbers of campylobacters in water. *J. Hyg.* 89(2):185-190.
- Brennhovd, O., G. Kapperud, and G. Langeland. 1992. Survey of thermotolerant *Campylobacter* spp. and *Yersinia* spp. in three surface water sources in Norway. *Int. J. Food Microbiol.* 15:327-338.
- Brinez, W. J., A. X. Roig-Sagues, M. M. Hernandez Herrero, T. Lopez-Pedemonte, and B. Guamis. 2006. Bactericidal efficacy of peracetic acid in combination with hydrogen peroxide against pathogenic and non-pathogenic strains of *Staphylococcus* spp., *Listeria* spp., and *Escherichia coli*. *Food Contr.* 17(7):516-521.
- Brown, H. G. 2003. Microbial problems, causes, and solutions in meat and poultry processing operations. Pages 89-93 in the Proceedings of the 56th American Meat Science Association Reciprocal Meat Conference. Columbia, MI.
- Buhr, R. J., N. A. Cox, N. J. Stern, M. T. Musgrove, J. L. Wilson, and K. L. Hiatt. 2002. Recovery of *Campylobacter* from segments of the reproductive tract of broiler breeder hens. *Avian Dis.* 46(4):919-924.
- Buswell, C. M., Y. M. Herlihy, L. M. Lawrence, J. T. M. McGuiggan, P. D. Marsh, C. W. Keevil, and S. A. Leach. 1998. Extended survival and persistence of *Campylobacter* spp. in water and aquatic biofilms and their detection by immunofluorescent-antibody and rRNA staining. *Appl. Environ. Microbiol.* 64(2):733-741.
- Butzler, J. P. 2004. *Campylobacter*, from obscurity to celebrity. *Clin. Microbiol. Infect.* 10(10):868-876.

- Capita, R., C. Alonso-Calleja, M. C. Garcia-Fernandez, and B. Moreno. 2002. Review: trisodium phosphate (TSP) treatment for decontamination of poultry. *Food Sci. Technol. Int.* 8(1):11-24.
- Carciofi, B. A. M., and J. B. Laurindo. 2007. Water uptake by poultry carcasses during cooling by water immersion. *Chem. Eng. Process.* 46(5):444-450.
- Carmarda, A., D. G. Newell, R. Nasti, and G. Di Modugno. 2000. Genotyping *Campylobacter jejuni* strains isolated from the gut and oviduct of laying hens. *Avian Dis.* 44:907-912.
- Carroll, C. D., and C. Z. Alvarado. 2008. Comparison of air and immersion chilling on meat quality and shelf life of marinated broiler breast fillets. *Poult. Sci.* 87:368-372.
- Cason, J. A., N. A. Cox, R. J. Buhr, and L. J. Richardson. 2010. Comparison of the statistics of *Salmonella* testing of chilled broiler chicken carcasses by whole-carcass rinse and neck skin excision. *Poult. Sci.* 89(9):2038-2040.
- Cason, J. A., and A. Hinton, Jr. 2006. Coliforms, *Escherichia coli*, *Campylobacter* and *Salmonella* in a counterflow poultry scalding tank with a dip tank. *Int. J. Poult. Sci.* 5(9):846-849.
- Cason, J. A., A. Hinton, Jr., and K. D. Ingram. 2000. Coliform, *Escherichia coli*, and salmonellae concentrations in a multiple-tank, counterflow poultry scalding tank. *J. Food Prot.* 63(9):1184-1188.
- Centers for Disease Control and Prevention. 1993. Update: multistate outbreak of *Escherichia coli* O157:H7 infections from hamburgers, western United States, 1992-1993. *Morb. Mortal. Wkly Rep.* 42(14):258-263.

- Centers for Disease Control and Prevention. 2000. National Antimicrobial Resistance Monitoring System (NARMS) – 1999 annual report. <http://www.cdc.gov/narms>.
- Centers for Disease Control and Prevention. 2002. Preliminary FoodNet data on the incidence of infection with pathogens transmitted commonly through food - selected sites, United States, 2001. *Morb. Mortal. Wkly Rep.* 51(15):325-329.
- Chantarapanont, W., M. E. Berrang, and J. F. Frank. 2004. Direct microscopic observation of viability of *Campylobacter jejuni* on chicken skin treated with selected chemical sanitizing agents. *J. Food Prot.* 67(6):1146-1152.
- Chuma, T., T. Yamada, K. Yano, K. Okamoto, and H. Yugi. 1994. A survey of *Campylobacter jejuni* in broilers from assignment to slaughter using DNA-DNA hybridization. *J. Vet. Med. Sci.* 56(4):697-700.
- CIA World Factbook. 2017. <https://www.cia.gov/library/publications/the-world-factbook/>.
- Clarke, A. G., and D. H. Bueschkens. 1985. Laboratory infection of chicken eggs with *Campylobacter jejuni* by using temperature or pressure differentials. *Appl. Environ. Microbiol.* 49(6):1467-1471.
- Cloak, O. M., G. Duffy, J. J. Sheridan, I. S. Blair, and D. A. McDowell. 2001. A survey on the incidence of *Campylobacter* spp. and the development of a surface adhesion polymerase chain reaction (SA-PCR) assay for the detection of *Campylobacter jejuni* in retail meat products. *Food Microbiol.* 18(3):287-298.
- Conner, D. E., and S. F. Biligli. 1994. Skin attachment model for improved laboratory evaluation of potential carcass disinfectants for their efficacy against *Salmonella* attached to broiler skin. *J. Food Prot.* 57(8):684-688.

- Coppen, P. P. 1993. *Salmonella* reduction in poultry processing - application of TSP process to European industry. Pages 524-531 in Proceedings of the 11th European Symposium on the Quality of Poultry Meat. Branche Francaise.
- Corry, J. E., C. James, S. J. James, and M. Hinton. 1995. *Salmonella*, *Campylobacter* and *Escherichia coli* 0157:H7 decontamination techniques for the future. Int. J. Food Microbiol. 28(2):187-196.
- Cox, N. A., J. S. Bailey, L. J. Richardson, R. J. Buhr, D. E. Cosby, J. L. Wilson, K. L. Hiatt, G. R. Siragusa, and D. V. Bourassa. 2005b. Presence of naturally occurring *Campylobacter* and *Salmonella* in the mature and immature ovarian follicles of late-life broiler breeder hens. Avian Dis. 49(2):285-287.
- Cox, N. A., C. L. Hofacre, J. S. Bailey, R. J. Buhr, J. L. Wilson, K. L. Hiatt, L. J. Richardson, M. T. Musgrove, D. E. Cosby, J. D. Tankson, Y. L. Vizzier, P. F. Cray, L. E. Vaughn, P. S. Holt, and D. V. Bourassaa. 2005a. Presence of *Campylobacter jejuni* in various organs one hour, one day, and one week following oral or intracloacal inoculation of broiler chicks. Avian Dis. 49(1):155-158.
- Cox, N. A., L. J. Richardson, M. E. Berrang, P. J. Fedorka-Cray, and R. J. Buhr. 2009a. *Campylobacter coli* naturally resistant to elevated levels of gentamicin as a marker strain in poultry research. J. Food Prot. 72(6):1288-1292.
- Cox, N. A., L. J. Richardson, R. J. Buhr, and P. J. Fedorka-Cray. 2009b. *Campylobacter* species occurrence within internal organs and tissues of commercial caged leghorn laying hens. Poult. Sci. 88(11):2449-2456.
- Cox, N. A., L. J. Richardson, R. J. Buhr, P. J. Fedorka-Cray, J. S. Bailey, J. L. Wilson,

- and K. L. Hiatt. 2006. Natural presence of *Campylobacter* spp. in various internal organs or commercial broiler breeder hens. *Avian Dis.* 50(3):450-453.
- Cox, N. A., L. J. Richardson, R. J. Buhr, J. K. Northcutt, J. S. Bailey, P. J. Fedorka Cray, and K. L. Hiatt. 2007. Recovery of *Campylobacter* and *Salmonella* serovars from the spleen, liver/gallbladder, and ceca of six and eight week old commercial broilers. *J. Appl. Poult. Res.* 16(4):477-480.
- Cox, N. A., L. J. Richardson, J. A. Cason, R. J. Buhr, Y. Vizzier-Thaxton, D. P. Smith, P. J. Fedorka-Cray, C. P. Romanenghi, L. V. Pereira, and M. P. Doyle. 2010. Comparison of neck skin excision and whole carcass rinse sampling methods for microbiological evaluation of broiler carcasses before and after immersion chilling. *J. Food Prot.* 73(5):976-980.
- Cox, N. A., N. J. Stern, J. L. Wilson, M. T. Musgrove, R. J. Buhr, and K. L. Hiatt. 2002. Isolation of *Campylobacter* spp. from semen samples of commercial broiler breeder roosters. *Avian Dis.* 46(3):717-720.
- Curtis, P., and J. Butler. 2009. Controlling *Campylobacter* in poultry plants. Auburn University. <http://fsis.usda.gov/>.
- Davies, A., and R. Board. 1998. The microbiology of meat and poultry. 1st ed. A. Davis and R. Board, eds. Blackie Academic and Professional. London, UK.
- Demirok, E., G. Veluz, W. V. Stuyvenberg, M. P. Castaneda, A. Byrd, and C. Z. Alvarado. 2013. Quality and safety of broiler meat in various chilling systems. *Poult. Sci.* 92(4):1117-1126.
- Deumier, F. 2004. Decontamination of deboned chicken legs by vacuum tumbling in lactic acid solution. *Int. J. Food Sci. Tech.* 41(1):23-32.

- DeWaal, C. S., C. Roberts, and C. Catella. 2012. Antibiotic resistance in foodborne pathogens: evidence of the need for risk management strategy. Center for Science in the Public Interest, White Paper. <http://cspinet.org/new/pdf/abrfoodbornenepathogenswhitepaper.pdf>.
- Dickens, J. A., B. G. Lyon, A. D. Whittemore, and C. E. Lyon. 1994. The effect of an acetic acid dip on carcass appearance, microbiological quality, and cooked breast meat texture and flavor. *Poult. Sci.* 73(4):576-581.
- Dickens, J. A., and A. D. Whittemore. 1995. The effects of extended chilling times with acetic acid on the temperature and microbiological quality of processed poultry carcasses. *Poult. Sci.* 74(6):1044-1048.
- Dickson, J. S., and M. E. Anderson. 1992. Microbiological decontamination of food animal carcasses by washing and sanitizing systems: a review. *J. Food Prot.* 55(2):133-140.
- Dincer, A. H., and A. Unluturk. 2001. Effects of lactic acid and fumaric acids on microbiological stability of turkey breast meat. IX European Symposium on the quality of eggs and egg products and XV European symposium on the quality of poultry meat. Symposium Book.
- Djenane, D., A. Sanchez-Escalante, J. A. Beltran, and P. Roncales. 2003. The shelf life of beef steaks treated with DL-lactic acid and antioxidants and stored under modified atmospheres. *Food Microbiol.* 20(1):1-7.
- Drewniak, E. E., M. A. Howe, Jr., J. E. Goresline, and E. R. Baush. 1954. Studies on sanitizing methods for use in poultry processing. Circular no. 930. U.S. Department of Agriculture. Washington, D.C.

- Dreyfuss, M. S., G. M. Ransom, M. D. Russell, K. E. Barlow, K. M. Pritchard, D. R. Eblen, C. A. Nadon, P. K. Saini, N. D. O. Antoine, B. E. Rose, and G. W. Zirnstein. 2007. Pathogen control in meat and poultry production: implementing the USDA food safety and inspection service's hazard analysis and critical control point system. *Infectious Disease: Foodborne Dis.* 15:383-404.
- Dubois-Brissonnet, F. 2012. Adaptation of *Salmonella* to antimicrobials in food processing environments. Pages 123-146 in *Salmonella: Distribution, Adaptation, Control Measures and Molecular Technologies*. B. A. Annous, and J. B. Gurtler, eds. Intech.
- Ellerbroek, L., E. M. Okolocha, and E. Weise. 1996. Lactic acid and trisodium phosphate for decontamination of poultry meat. Pages 187-195 in *Microbial Methods for the Meat Industry*. M. H. Hinton, and C. Rowlings, eds. University of Bristol Press. Bristol.
- Engberg, J., F. M. Aarestrup, D. E. Taylor, P. Gerner-Smidt, and I. Nachamkin. 2001. Quinolone and macrolide resistance in *Campylobacter jejuni* and *C. coli*: Resistance mechanisms and trends in human isolates. *Emerg. Infect. Dis.* 7(1):24-34.
- Evans, S. J., and A. R. Sayers. 2000. A longitudinal study of *Campylobacter* infection of broiler flocks in Great Britain. *Prev. Vet. Med.* 46(3):209-223.
- FAO. 2003. Risk assessment of *Campylobacter* spp. in broiler chickens and *Vibrio* spp. in seafood. Report of a Joint FAO/WHO Expert Consultation. Bangkok, Thailand.
- FAO [Food and Agriculture Organization of the United Nations]. 2009. *Salmonella* and

- Campylobacter* in chicken meat: meeting report. Page 56 in Microbiological Risk Assessment Series No. 19. Rome.
- FDA. Federal Register. 2000. Secondary direct additives permitted in food for human consumption. Peroxyacids – Final Rule, 21 CFR Part 173. 65(228):70660-70661.
- FDA. 2015. Food Protection Plan of 2007. <https://www.fda.gov/Food/GuidanceRegulation/FoodProtectionPlan207.default.htm>.
- Federal Register, United States Department of Agriculture. 1982. Meat and poultry products: phosphates and sodium hydroxide. Fed. Regis. 47:10779.
- Federal Register, United States Department of Agriculture. 1996. 9 CFR Part 304 et al., Pathogen Reduction; Hazard Analysis and Critical Control Point (HACCP) Systems. Final Rule. 61(144):38846-38848.
- Ferrara, O. 2005. Brand preferences among meats: an application of probit models. University of Florida. Master of Science Thesis.
- Fletcher, D. L. 2002. Poultry meat quality. World's Poult Sci. J. 58(2):131-145.
- Fluckey, W. M., M. X. Sanchez, S. R. Mckee, D. Smith, E. Pendleton, and M. M. Brashears. 2003. Establishment of a microbiological profile for an air-chilling poultry operation in the United States. J. Food Prot. 66(2):272-279.
- Fortin, N. D. 2003. The hang-up with HACCP: the resistance to translating science into food safety law. Food Drug Law J. 58(4):565-593.
- Friedman, C. R., J. Neimann, H. C. Wegener, and R. V. Tauxe. 2000. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. Pages 121–138 in American Society for Microbiology Press. I. Nachamkin, and M. J. Blaser, eds. Washington, D.C.

- Gallo, L., R. E. Schmitt, and W. Schmidt-Lorenz. 1988. Microbial spoilage of refrigerated fresh broilers: I. Bacterial flora and growth during storage. *Food Sci. Tech.* 21(4):216-223.
- Gaudreau, C., and H. Gilbert. 1998. Antimicrobial resistance of clinical strains of *Campylobacter jejuni* subsp. *jejuni* isolated from 1985 to 1997 in Quebec, Canada. *Antimicrob. Agents Chemother.* 42(8):2106-2108.
- Gavin, A., and L. M. Weddig. 1995. Canned foods: principles of thermal process control, acidification, and container closure evaluation. Pages 36-41 in the *Food Processors Institute*.
- Ge, B., D. G. White, P. F. McDermott, W. Girand, S. Zhao, S. Hubert, and J. Meng. 2003. Antimicrobial-resistant *Campylobacter* species from retail raw meats. *Appl. Environ. Microbiol.* 69(5):3005-3007.
- Geornaras, I., and A. von Holy. 2000. Bacterial counts associated with poultry processing at different sampling times. *J. Basic Microbiol.* 40:343-349.
- Geornaras, I., A. de Jesus, E. van Zyl, and A. von Holy. 1995. Microbiological survey of a South African poultry processing plant. *J. Basic Microbiol.* 35(2):73-82.
- Gibbens, J. C., S. J. Pascoe, S. J. Evans, R. H. Davies, and A. R. Sayers. 2001. A trial of biosecurity as a means to control *Campylobacter* infection of broiler chickens. *Prev. Vet. Med.* 48(2):85-99.
- Giese, J. 1992. Experimental process reduces *Salmonella* on poultry. *Food Technol.* 46:112.
- Giese, J. 1993. *Salmonella* reduction process receives approval. *Food Technol.* 47:110.
- Gilbert, C. D., Y. Bai, and H. Jiang. 2015. Microbial evaluation of Cecure treated (post

- chill) raw poultry carcasses and cut-up parts in four commercial broiler processing facilities. *Int. J. Poult. Sci.* 14(3):120-126.
- Gill, C. O., and M. Badoni. 2004. Effects of peroxyacetic acid, acidified sodium chlorite or lactic acid solutions on the microflora of chilled beef carcasses. *Int. J. Food Microbiol.* 91(1):43-50.
- Gonzalez-Fandos, E., B. Herrera, and N. Maya. 2009. Efficacy of citric acid against *Listeria monocytogenes* attached to poultry skin during refrigerated storage. *Int. J. Food Sci. Technol.* 44(2):262-268.
- Gregory, E., H. Barnhart, D. W. Dreesen, N. J. Stern, and J. L. Corn. 1997. Epidemiological study of *Campylobacter* spp. in broilers: source, time of colonization, and prevalence. *Avian Dis.* 41(4):890-898.
- Gupta, A., J. M. Nelson, T. J. Barrett, R. V. Taxue, S. P. Rossiter, C. R. Friedman, K. W. Joyce, K. E. Smith, T. F. Jones, M. A. Hawkins, B. Shiferaw, J. L. Beebe, D. J. Vugia, T. Rabatsky-Ehr, J.A. Benson, T. P. Root, and F. J. Angulo. 2004. Antimicrobial resistance among *Campylobacter* strains, United States, 1997-2001. *Emerg. Infect. Dis.* 10(6):1102-1109.
- Hald, B., A. Wedderkopp, and M. Madsen. 2000. Thermophilic *Campylobacter* spp. in Danish broiler production: a cross-sectional survey and a retrospective analysis of risk factors for occurrence in broiler flocks. *Avian Pathol.* 29(2):123-131.
- Harris, N. V., D. Thompson, D. C. Martin, and C. M. Nolan. 1986. A survey of *Campylobacter* and other bacterial contaminants of pre-market chicken and retail poultry and meats, King County, Washington. *Am. J. Public Health.* 76(4):401-406.

- Harrison, W. A., C. J. Griffith, D. Tennant, and A. C. Peters. 2001. Incidence of *Campylobacter* and *Salmonella* isolated from retail chicken and associated packaging in South Wales. *Lett. Appl. Microbiol.* 33(6):450-454.
- Hiett, K. L., N. J. Stern, P. Fedorka-Cray, N. A. Cox, M. T. Musgrove, and S. Ladely. 2002. Molecular subtype analyses of *Campylobacter* spp. from Arkansas and California poultry operations. *Appl. Environ. Microbiol.* 68(12):6220-6236.
- Havenstein, G. B. 2006. Performance changes in poultry and livestock following 50 years of genetic selection. *Lohmann Info.* 41:30-37.
- Hinton Jr., A., J. A. Cason, M. E. Hume, and K. D. Ingram. 2004a. Use of MIDI-fatty acid methyl ester analysis to monitor the transmission of *Campylobacter* during commercial poultry processing. *J. Food Prot.* 67(8):1610-1616.
- Hinton Jr., A., J. A. Cason, M. E. Hume, and K. D. Ingram. 2004b. Spread of *Campylobacter* spp. during poultry processing in different seasons. *Int. J. Poult. Sci.* 3(7):432-437.
- Hinton Jr., A., J. A. Cason, and K. D. Ingram. 2002. Enumeration and identification of yeasts associated with commercial poultry processing and spoilage of refrigerated broiler carcasses. *J. Food Prot.* 65(6):993-998.
- Hinton Jr., A., J. A. Cason, and K. D. Ingram. 2004c. Tracking spoilage bacteria in commercial poultry processing and refrigerated storage of poultry carcasses. *Int. J. Food Microbiol.* 91(2):155-165.
- Hudson, J. A., C. Nicol, J. Wright, R. Whyte, and S. K. Hasell. 1999. Seasonal variation of *Campylobacter* types from human cases, veterinary cases, raw chicken, milk and water. *J. Appl. Microbiol.* 87(1):115-124.

- Hue, O., V. Allain, M. J. Laisney, S. Le Bouquin, F. Lalande, I. Petetin, S. Rouxel, S. Quesne, P. Y. Gloaguen, M. Picherot, J. Santolini, S. Bougeard, G. Salvat, and M. Chemaly. 2011. *Campylobacter* contamination of broiler caeca and carcasses at the slaughterhouse and correlation with *Salmonella* contamination. Food Microbiol. 28(5):862-868.
- Huezo, R., D. P. Smith, J. K. Northcutt, and D. L. Fletcher. 2007. Effect of immersion or dry air chilling on broiler carcass moisture retention and breast fillet functionality. J. Appl. Poult. Res. 16(3):438-447.
- Huffman, R. D. 2002. Current and future technologies for the decontamination of carcasses and fresh meat. Meat Sci. 62:285-294.
- Hui, Y. H., and F. Sherkat. 2005. Biosynthesis of natural aroma compounds. Page 193 in Handbook of Food Science, Technology, and Engineering. CRC Press. London, N.Y.
- Humphrey, T., S. O'Brien, and M. Madsen. 2007. Campylobacters as zoonotic pathogens: a food production perspective. Int. J. Food Micro. 117(3):237-257.
- Hutchinson, M. L., L. D. Walters, G. C. Mead, M. Howell, and V. M. Allen. 2006. An assessment of sampling methods and microbiological hygiene indicators for process verification in poultry slaughterhouses. J. Food Prot. 69(1):145-153.
- Immerseel, F. V., L. De Zutter, K. Houf, F. Pasmans, F. Haesebrouck, and R. Ducatelle. 2009. Strategies to control *Salmonella* in broiler production chain. World Poult. Sci. J. 65(3):367-392.
- Inspection Methods. 2018. Pathogen reduction - *Salmonella* and *Campylobacter*

- performance standards verification testing. Food Safety Inspection Service (FSIS). <https://www.fsis.usda.gov/>.
- International Commission and Microbiological Specifications for Foods (ICMSF). 1998. Microbiological ecology of food commodities. Pages 107-157 in Poultry and Poultry Products. Blackie Academic and Professional. London, UK.
- Izat, A. L., M. Colberg, R. A. Thomas, M. H. Adams, and C. D. Driggers. 1990. Effects of lactic acid in processing waters on the incidence of salmonellae on broilers. *J. Food Qual.* 13(4):295-306.
- Izat, A. L., F. A. Gardner, J. H. Denton, and F. A. Golan. 1988. Incidence and level of *Campylobacter jejuni* in broiler processing. *Poult. Sci.* 67(11):1568-1572.
- Jacobs-Reitsma, W. F. 1997. Aspects of epidemiology of *Campylobacter* in poultry. *Vet. Q.* 19(3):113-117.
- Jacobs-Reitsma, W. F., N. M. Bolder, and R. W. A. W. Mulder. 1994. Cecal carriage of *Campylobacter* and *Salmonella* in Dutch broiler flocks at slaughter: a one-year study. *Poult. Sci.* 73(8):1260-1266.
- Jacobs-Reitsma, W. F., A. W. van de Giessen, N. M. Bolder, and R. W. Mulder. 1995. Epidemiology of *Campylobacter* spp. at two Dutch broiler farms. *Epidemiol. Infect.* 114(3):413-421.
- James, C., C. Vincent, T. I. de Andrade Lima, and S. J. James. 2006. The primary chilling of poultry carcasses - a review. *Int. J. Refrig.* 29(6):847-862.
- Jones, D. M., E. M. Sutcliffe, and A. Curry. 1991. Recovery of viable but non-culturable *Campylobacter jejuni*. *J. Gen. Microbiol.* 137(10):2477-2482.
- Kapperud, G., E. Skjerve, L. Vik, K. Hauge, A. Lysaker, I. Aalmen, S. M. Ostroff, and

- M. Potter. 1993. Epidemiological investigation of risk factors for *Campylobacter* colonization in Norwegian broiler flocks. *Epidemiol. Infect.* 111(2):245-255.
- Keenan, S., S. Spice, J. Cole, and P. Banfi. 2015. Food safety policy and regulation in the United States. In: European Parliament's Committee on Environment, Public Health and Food Safety (ENVI). Brussels, Belgium. [https://www.europarl.europa.eu/RegData/etudes/STUD/2015/536324/IPOL_STU\(2015\)536324_EN.pdf](https://www.europarl.europa.eu/RegData/etudes/STUD/2015/536324/IPOL_STU(2015)536324_EN.pdf).
- Keener, K. M., M. P. Bashor, P. A. Curtis, B. W. Sheldon, and S. Kathariou. 2004. Comprehensive review of *Campylobacter* and poultry processing. *Compr. Rev. Food Sci. Food Saf.* 3(2):105-116.
- Kemp, G. K., M. L. Aldrich, M. L. Guerra, and K. R. Schneider. 2001. Continuous online processing of fecal- and ingesta- contaminated poultry carcasses using an acidified sodium chlorite antimicrobial intervention. *J. Food Prot.* 64(6):807-812.
- Kemp, G. K., M. L. Aldrich, and A. L. Waldroup. 2000. Acidified sodium chlorite antimicrobial treatment of broiler carcasses. *J. Food Prot.* 63(8):1087-1092.
- Kessel, A. S., I. A. Gillespie, S. J. O'Brien, G. K. Adak, T. J. Humphrey, and L. R. Ward. 2001. General outbreaks of infectious intestinal disease linked with poultry, England and Wales, 1992-1999. *Commun. Dis. Public Health.* 4(3):171-177.
- Kim, J. W., S. J. Knabel, and S. Doores. 1993. Penetration of *Salmonella* Typhimurium into turkey skin. *J. Food Prot.* 56(4):292-296.
- Kim, J. W., M. F. Slavik, F.G. Bender. 1994. Removal of *Salmonella* Typhimurium attached to chicken skin by rinsing with trisodium phosphate solution: scanning electron microscopic examination. *J. Food Safety.* 14(1):77-84.
- Korber, D. R., A. Choi, G. M. Wolfaardt, and D. E. Caldwell. 1996. Bacterial

- plasmolysis as a physical indicator of viability. *Appl. Environ. Microbiol.* 62(11):3939-3947.
- Lastovica, A. J. 2006. Emerging *Campylobacter* spp.: the top of the iceberg. *Clin. Microbiol. Newsletter.* 28(7):49-56.
- Lecompte, J. Y., A. Collignan, S. Sarter, E. Cardinale, and A. Kondjoyan. 2009. Decontamination of chicken surfaces inoculated with *Listeria innocua*, *Salmonella* Enteritidis, and *Campylobacter jejuni* by contact with a concentrated lactic acid solution. *Br. Poult. Sci.* 50(3):307-317.
- Lee, M. D., S. Sanchez, M. Zimmer, U. Idris, M. E. Berrang, and P. F. McDermott. 2002. Class 1 Integron-associated tobramycin-gentamicin resistance in *Campylobacter jejuni* isolated from the broiler chicken house environment. *Antimicrob. Agents Chemother.* 46(11):3660-3664.
- Leistner, L., and L. G. M. Gorris. 1995. Food preservation by hurdle technology. *Trends Food Sci. Technol.* 6(2):41-46.
- Li, Y., M. F. Slavik, J. T. Walker, and H. Xiong. 1997. Pre-chill spray of chicken carcasses to reduce *Salmonella* Typhimurium. *J. Food Sci.* 62(3):605-607.
- Liem, D. 2002. Personal communication. Environmental Protection Agency: Office of Pesticide Programs, Antimicrobial Division. Washington, D.C.
- Lillard, H. S. 1979. Levels of chlorine and chlorine dioxide of equivalent bactericidal effect in poultry processing water. *J. Food Sci.* 44(6):1594-1597.
- Lillard, H. S. 1980. Effect on broiler carcasses and water of treating chiller water with chlorine or chlorine dioxide. *Poult. Sci.* 59(8):1761-1766.
- Lillard, H. S. 1985. Bacterial cell characteristics and conditions influencing their

- adhesion to poultry skin. *J. Food Prot.* 48(9):803-807.
- Lillard, H. S. 1986. Distribution of “attached” *Salmonella* Typhimurium cells between poultry skin and a surface film following water immersion. *J. Food Prot.* 49(6):449-454.
- Lillard, H. S. 1988. Effect of surfactant or changes in ionic strength on the attachment of *Salmonella* Typhimurium to poultry skin and muscle. *J. Food Sci.* 53(3):727-730.
- Lillard, H. S. 1989. Incidence and recovery of salmonellae and other bacteria from commercially processed poultry carcasses at selected pre- and post-evisceration steps. *J. Food Prot.* 52(2):88-91.
- Loretz, M., R. Stephan, and C. Zweifel. 2010. Antimicrobial activity of decontamination treatments for poultry carcasses: a literature survey. *Food Control.* 21(6):791-804.
- Man, S. M. 2011. The clinical importance of emerging *Campylobacter* species. *Nat. Rev. Gastroenterol. Hepatol.* 8(12):669-685.
- Mbata, T. 2017. Poultry meat pathogens and its control. *Int. J. Food Safety.* 7:20-28.
- McCarthy, N., and J. Giesecke. 2001. Incidence of Guillain-Barre Syndrome following infection with *Campylobacter jejuni*. *Am. J. Epidemiol.* 153(6):610-614.
- McKee, S. 2001. Chilling of poultry, immersion and air. *Watt Poultry USA.* 12:18-24.
- McMeekin, T. A., and C. J. Thomas. 1978. Retention of bacteria on chicken skin after immersion in bacterial suspensions. *J. Appl. Bacteriol.* 45(3):383-387.
- Mead, G. C., V. M. Allen, C. H. Burton, and J. E. L. Corry. 2000. Microbial cross contamination during air chilling of poultry. *Br. Poult. Sci.* 41(2):158-162.
- Mead, G. C., W. R. Hudson, and M. H. Hinton. 1995. Effect of changes in processing to

- improve hygiene control on contamination of poultry carcasses with *Campylobacter*. *Epidemiol. Infect.* 115(3):495-500.
- Mead, P. S., L. Slutsker, V. Dietz, L. F. McCaig, J. S. Bresee, C. Shapiro, P. M. Griffin and R. V. Tauxe. 1999. Food-related illness and death in the United States. *Emerg. Infect. Dis.* 5(5):607-625.
- Mehyar, G. F., G. Blank, J. H. Han, A. Hydamaka, and R. A. Holley. 2005. Effectiveness of trisodium phosphate, lactic acid and commercial antimicrobials against pathogenic bacteria on chicken skin. *Food Prot. Trends.* 25:351-362.
- Meinersmann, R. J., W. E. Rigsby, N. J. Stern, L. C. Kelley, J. E. Hill, and M. P. Doyle. 1991. Comparative study of colonizing and noncolonizing *Campylobacter jejuni*. *Am. J. Vet. Res.* 52(9):1518-1522.
- Moore, J. E., M. D. Barton, I. S. Blair, D. Corcoran, J. S. Dooley, S. Fanning, I. Kempf, A. J. Lastovica, C. J. Lowery, M. Matsuda, D. A. McDowell, A. McMahon, B. C. Millar, J. R. Rao, P. J. Rooney, B. S. Seal, W. J. Snelling, and O. Tolba. 2006. The epidemiology of antibiotic resistance in *Campylobacter*. *Microbes Infect.* 8(7):1955-1966.
- Moore, J. E., M. Crowe, N. Heaney, and E. Crothers. 2001. Antibiotic resistance in *Campylobacter* spp. isolated from human feces (1980-2000) and foods (1997-2000) in Northern Ireland: an update. *J. Antimicrob. Chemother.* 48(3):455-457.
- Moore, J. E., T. S. Wilson, D. R. Wareing, T. J. Humphrey, and P. G. Murphy. 2002. Prevalence of thermophilic *Campylobacter* spp. in ready-to-eat foods and raw poultry in Northern Ireland. *J. Food Prot.* 65(8):1326-1328.
- Moran, A. P., and M. E. Upton. 1986. A comparative study of the rod and coccoid forms

- of *Campylobacter jejuni* ATCC 29428. J. Appl. Bacteriol. 60(2):103-110.
- Mulder, R. W. A. W., L. W. J. Dorresteijn, and J. van der Broek. 1978. Cross contamination during the scalding and plucking of broilers. Br. Poult. Sci. 19:61-70.
- Mulder, R. W. A. W., M. C. van der Hulst, and N. M. Bolder. 1987. *Salmonella* decontamination of broiler carcasses with lactic acid, l-cysteine, and hydrogen peroxide. Poult. Sci. 66:1555-1557.
- Murphy, C., C. Carroll, and K. N. Jordan. 2003. Induction of an adaptive tolerance response in the foodborne pathogen, *Campylobacter jejuni*. FEMS Microbiol. Lett. 223(1):89-93.
- Murphy, C., C. Carroll, and K. N. Jordan. 2006. Environmental survival mechanisms of the foodborne pathogen *Campylobacter jejuni*. J. Appl. Microbiol. 100(4):623-632.
- Nachamkin, I. 1995. *Campylobacter* and *Arcobacter*. Pages 483-491 in Manual of Clinical Microbiology, 6th ed. P. R. Murray, E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. J. Tenover, eds. Washington, D.C.
- Nachamkin, I., B. M. Allos, and T. Ho. 1998. *Campylobacter* species and Guillain-Barre syndrome. Clin. Microbiol. Rev. 11(3):555-567.
- Nadeau, E., S. Messier, and S. Quessy. 2002. Prevalence and comparison of genetic profiles of *Campylobacter* strains isolated from poultry and sporadic cases of campylobacteriosis in humans. J. Food Prot. 65(1):73-78.
- Nagel, G. M., L. J. Bauermeister, C. L. Bratcher, M. Singh, and S. R. McKee. 2013.

- Salmonella* and *Campylobacter* reduction and quality characteristics of poultry carcasses treated with various antimicrobials in a post-chill immersion tank. *Int. J. Food Microbiol.* 165(3):281-286.
- Nauta, M. J., A. Hill, H. Rosenquist, S. Brynestad, A. Fetsch, P. van der Logt, A. Fazil, B. B. Christensen, E. Katsma, B. Borck, and A. H. Havelaar. 2009. A comparison of risk assessments on *Campylobacter* in broiler meat. *Int. J. Food Microbiol.* 129:107-123.
- National Antimicrobial Resistance Monitoring System (NARMS). 2015. NARMS integrated report: 2012-2013. Food and Drug Administration.
- National Chicken Council (NCC). 2018. Broiler chicken industry key facts 2018. <https://www.nationalchickencouncil.org/about-the-industry/statistics/broiler-chicken-industry-key-facts/>.
- Nelson, J. M., T. M. Chiller, J. H. Powers, and F. J. Angulo. 2007. Fluoroquinolone resistant *Campylobacter* species and the withdrawal of fluoroquinolones from use in poultry: a public health success story. *Clin. Infect. Dis.* 44(7):977-980.
- Newell, D. G., and H. C. Davison. 2003. *Campylobacter* - control and prevention. Page 470 in Chapter 22: Microbial Food Safety in Animal Agriculture. M. E. Torrence, and R. E. Isaacson, eds. Wiley-Blackwell Publishing.
- Newell, D. G., and J. A. Wagenaar. 2000. Poultry infections and their control at the farm level. Pages 497-510 in *Campylobacter*, 2nd ed. I. Nachamkin, M. J. Blaser, eds. Washington, D.C.
- Northcutt, J. K., M. E. Berrang, J. A. Dickens, D. L. Fletcher, and N. A. Cox. 2003a.

- Effect of broiler age, feed withdrawal, and transportation on levels of coliforms, *Campylobacter*, *Escherichia coli* and *Salmonella* on carcasses before and after immersion chilling. *Poult. Sci.* 82(1):169-173.
- Northcutt, J. K., R. J. Buhr, M. E. Berrang, and D. L. Fletcher. 2003b. Effects of replacement finisher feed and length of feed withdrawal on broiler carcass yield and bacteria recovery. *Poult. Sci.* 82(11):1820-1824.
- Northcutt, J. K., and S. M. Russell. 2010. General guidelines for implementation of HACCP in a poultry processing plant. The cooperative extension, University of Georgia. <http://www.caes.uga.edu/>.
- Northcutt, J. K., D. Smith, R. I. Huezo, and K. D. Ingram. 2008. Microbiology of broiler carcasses and chemistry of chiller water as affected by water reuse. *Poult. Sci.* 87(7):1458-1463.
- Northcutt, J. K., D. P. Smith, M. T. Musgrove, K. D. Ingram, and A. Hinton, Jr. 2005. Microbiological impact of spray washing broiler carcasses using different chlorine concentrations and water temperatures. *Poult. Sci.* 84(10):1648-1652.
- Oyarzabal, O. A. 2005. Reduction of *Campylobacter* spp. by commercial antimicrobials applied during the processing of broiler chickens: a review from the United States perspective. *J. Food Prot.* 68(8):1752-1760.
- Oyarzabal, O. A., C. Hawk, S. F. Bilgili, C. C. Warf, and G. K. Kemp. 2004. Effects of post-chill application of acidified sodium chlorite to control *Campylobacter* spp. and *Escherichia coli* on commercial broiler carcasses. *J. Food Prot.* 67(10):2288-2291.
- Pacholewicz, E., A. Swart, J. A. Wagenaar, L. J. Lipman, and A. H. Havelaar. 2016.

- Explanatory variables associated with *Campylobacter* and *Escherichia coli* concentrations on broiler chicken carcasses during processing in two slaughterhouses. *J. Food Prot.* 79(12):2038-2047.
- Padungtod, P., R. Hanson, D. L. Wilson, J. Bell, J. E. Linz, and J. B. Kaneene. 2002. Identification of *Campylobacter jejuni* isolates from cloacal and carcass swabs in chickens in Thailand by a 5' nuclease fluorogenic polymerase chain reaction assay. *J. Food Prot.* 65(11):1712-1716.
- Painter, J. A., R. M. Hoekstra, T. Ayers, R. V. Tauxe, C. R. Braden, F. J. Angulo, and P. M. Griffin. 2013. Attribution of foodborne illness, hospitalizations, and deaths to food commodities by using outbreak data, United States, 1998-2008. *Emerg. Infect. Dis.* 19(3):407-415.
- Park, S. F. 2002. The physiology of *Campylobacter* species and its relevance to their role as foodborne pathogens. *Int. J. Food Microbiol.* 74(3):177-188.
- Parveen, S., M. Taabodi, J. G. Schwarz, T. P. Oscar, J. Harter-Dennis, and D. G. White. 2007. Prevalence and antimicrobial resistance of *Salmonella* recovered from processed poultry. *J. Food Prot.* 70(11):2466-2472.
- Pearson, A. D., M. Greenwood, T. D. Healing, D. M. Rollins, M. Shahamat, J. Donaldson, and R. R. Colwell. 1993. Colonization of broiler chickens by waterborne *Campylobacter jejuni*. *Appl. Environ. Microbiol.* 59(4):987-996.
- Petracci, M., and C. Cavani. 2012. Muscle growth and poultry meat quality issues. *Nutrients.* 4(1):1-12.
- Petracci, M., S. Mudalal, E. Babini, and C. Cavani. 2014. Effect of white striping on chemical composition and nutritional value of chicken breast meat. *Ital.*

- J. Anim. Sci. 13:179-183.
- Petrak T., Z. Kalodera, P. Novakovic, and L. Gumhalter Karolyi. 1999. Bacteriological comparison of parallel and counter flow water chilling of poultry meat. *Meat Sci.* 53:269-271.
- PEW. 2016. Emerging pathogens in meat and poultry. Pages 1-73. PEW Charitable Trusts.
- Piddock, L. J. 1999. Mechanisms of fluoroquinolone resistance: an update 1994-1998. *Drugs.* 58(2):11-18.
- Rajkovic, A., N. Tomic, N. Smigic, M. Uyttendaele, P. Ragaert, and F. Devlieghere. 2010. Survival of *Campylobacter jejuni* on raw chicken legs packed in high-oxygen or high carbon dioxide atmosphere after the decontamination with lactic acid/sodium lactate buffer. *Int. J. Food Microbiol.* 140:201-206.
- Rasmussen, W. D. 1986. Lincoln's agricultural legacy. United States Department of Agriculture. <https://www.nal.usda.gov/lincolns-agricultural-legacy>.
- Rasschaert, G., V. Piessens, P. Scheldeman, S. Leleu, A. Stals, L. Herman, M. Heyndrickx, and W. Messens. 2013. Efficacy of electrolyzed oxidizing water and lactic acid on the reduction of *Campylobacter* on naturally contaminated broiler carcasses during processing. *Poult. Sci.* 92(4):1077-1084.
- Richardson, L. J., N. A. Cox, R. J. Buhr, and M. A. Harrison. 2011. Isolation of *Campylobacter* from circulating blood of commercial broilers. *Avian Dis.* 55(3):375-378.
- Ricke, S. C. 2014. Application of molecular approaches for understanding foodborne

- Salmonella* establishment in poultry production. Pages 1-25 in Advances in Biology. Hindawi Publishing Corporation. Wiley and Sons, Inc.
- Ricke, S. C., E. J. van Loo, M. G. Johnson, and C. A. O'Bryan. 2012. Alternatives to traditional antimicrobials for organically processed meat and poultry. Pages 211-216 in Organic Meat Production and Processing.
- Ridley, A. M., V. M. Allen, M. Sharma, J. A. Harris, and D. G. Newell. 2008. Real-time PCR approach for detection of environmental sources of *Campylobacter* strains colonizing broiler flocks. Appl. Environ. Microbiol. 74(8):2492-2504.
- Riedel, C. T., L. Brondsted, H. Rosenquist, S. N. Haxgart, and B. B. Christensen. 2009. Chemical decontamination of *Campylobacter jejuni* on chicken skin and meat. J. Food Prot. 72(6):1173-1180.
- Rio, E., M. Panizo-Moran, M. Prieto, C. Alonso-Calleja, and R. Capita. 2007. Effect of various chemical decontamination treatments on natural microflora and sensory characteristics of poultry. Int. J. Food Microbiol. 115:268-280.
- Robinson, D. A. 1981. Infective dose of *Campylobacter jejuni* in milk. Br. Med. J. 282:1584.
- Rollins, D. M., and R. R. Colwell. 1986. Viable but nonculturable stage of *Campylobacter jejuni* and its role in survival in the natural aquatic environment. Appl. Environ. Microbiol. 52(3):531-538.
- Rosenquist, H., L. Boysen, C. Galliano, S. Nordentoft, S. Ethelberg, and B. Borck. 2009. Danish strategies to control *Campylobacter* in broilers and broiler meat: facts and effects. Epidemiol. Infect. 137(12):1742-1750.
- Rouger, A., O. Tresse, and M. Zagorec. 2017. Bacterial contaminants of poultry meat:

- sources, species, and dynamics. *Microorganisms*. 50(5):1-16.
- Russell, S. M. 2007. *Salmonella* reduction at scalding and picking: a big opportunity. <http://www.wattpoultryusa-digital.com/PoultryUSA/Article.aspx?io=1290>.
- Russell, S. M. 2012a. Verification of the efficacy of intervention strategies. Pages 243-257 in *Controlling Salmonella in poultry production and processing*. Boca Raton. CRC Press. London, N.Y.
- Russell, S. M. 2012b. *Salmonella* intervention strategies and testing methods differ greatly between the United States and Europe. Pages 259-267 in *Controlling Salmonella in poultry production and processing*. Boca Raton. CRC Press. London, N.Y.
- Russell, S. M. 2012c. Intervention strategies for reducing *Salmonella* prevalence on ready to-cook chicken. *Cooperative Extension. Bulletin 1222*:1-13.
- Russell, S. M., D. L. Fletcher, and N. A. Cox. 1996. Spoilage bacteria of fresh broiler chicken carcasses. *Poult. Sci.* 74(12):2041-2047.
- Saenz, Y., M. Zarazaga, M. Lantero, M. J. Gastanares, F. Baquero, and C. Torres. 2000. Antibiotic resistance in *Campylobacter* strains isolated from animals, foods, and humans in Spain in 1997-1998. *Antimicrob. Agents Chemother.* 44(2):257-271.
- Saleha, A. A., G. C. Mead, A. L. Mead, and A. L. Ibrahim. 1998. *Campylobacter jejuni* in poultry production and processing in relation to public health. *J. World Poult. Sci.* 54(1):49-58.
- Sampathkumar, B., G. G. Khachatourians, and D. R. Korber. 2004. Treatment of

- Salmonella enterica* serovar Enteritidis with a sublethal concentration of trisodium phosphate or alkaline pH induces thermotolerance. *Appl. Environ. Microbiol.* 70(8):4613-4620.
- Sams, A. R. 2001. First processing: slaughter through chilling. Pages 19-34 in *Poultry Meat Processing*. A. R. Sams, ed. CRC Press. Boca Raton, FL.
- Sams, A. R., and C. Z. Alvarado. 2010. Introduction to poultry meat processing. Pages 1-3 in *Poultry Meat Processing*. C. M. Owens, C. Z. Alvarado, and A. R. Sams, eds. CRC Press. Boca Raton, FL.
- Sanchez, M. X., W. M. Fluckey, M. M. Brashears, and S. R. McKee. 2002. Microbial profile and antibiotic susceptibility of *Campylobacter* spp. and *Salmonella* spp. in broilers processed in air chilled and immersion chilled environments. *J. Food Prot.* 65(6):948-956.
- Scallan, E., P. M. Griffin, F. J. Angulo, R. V. Tauxe, and R. M. Hoekstra. 2011a. Foodborne illness acquired in the United States - unspecified agents. *Emerg. Infect. Dis.* 17(1):16-22.
- Scallan, E., R. M. Hoekstra, F. J. Angulo, R. V. Tauxe, M. Widdowson, S. L. Roy, J. L. Jones, and P. M. Griffin. 2011b. Foodborne illness acquired in the United States - major pathogens. *Emerg. Infect. Dis.* 17(1):7-15.
- Schreurs, F. J. G. 2000. Post mortem changes in chicken muscle. *World's Poult. Sci. J.* 56:319-346.
- Scientific Committee on Veterinary Measures Relating to Public Health (SCVMRPH).

1998. Benefits and limitations of antimicrobial treatments for poultry carcasses. European Commission. Brussels, Belgium. https://ec.europa.eu/food/sites/food/files/safety/docs/sci-com_scv_out14_en.pdf.
- Sellars, M. J., S. J. Hall, and D. J. Kelly. 2002. Growth of *Campylobacter jejuni* supported by respiration of fumarate, nitrate, nitrite, trimethylamine-*N*-oxide, or dimethyl sulfoxide requires oxygen. *J. Bacteriol.* 184(15):4187-4196.
- Shakil, S., R. Khan, R. Zarrilli, and A. U. Khan. 2008. Aminoglycosides versus bacteria—a description of the action, resistance mechanism, and nosocomial battleground. *J. Biomed. Sci.* 15(1):5-14.
- Skirrow, M. B. 1977. *Campylobacter* enteritis: a new disease. *Br. Med. J.* 2:9-11.
- Smith, R. N., R. N. Andersen, and P. E. Kolenbrander. 1991. Inhibition of intergeneric coaggregation among oral bacteria by cetylpyridinium chloride, chlorhexidine digluconate and octenidine dihydrochloride. *J. Periodontal Res.* 26(5):422-428.
- Smith, K. E., J. M. Besser, C. W. Hedberg, F. T. Leano, J. B. Bender, J. H. Wicklund, B. P. Johnson, K. A. Moore, and M. T. Osterholm. 1999. Quinolone-resistant *Campylobacter jejuni* infections in Minnesota, 1992-1998. *N. Engl. J. Med.* 340(20):1525-1532.
- Smulders, F. 1995. Preservation by microbial decontamination; the surface treatment of meats by organic acids. Pages 253-282 in *New Methods of Food Preservation*. G. W. Gould, ed. Blackie Academic and Professional, London.
- Stopforth, J. D., R. O'Connor, M. Lopes, B. Kottapalli, W. E. Hill, and M. Samadpour.

2007. Validation of individual and multiple-sequential interventions for reduction of microbial populations during processing of poultry carcasses and parts. *J. Food Prot.* 70(6):1393-1401.
- Talsma, E., W. G. Goettsch, H. L. Nieste, P. M. Schrijnemakers, and M. J. Sprenger. 1999. Resistance in *Campylobacter* species: increased resistance to fluoroquinolones and seasonal variation. *Clin. Infect. Dis.* 29(4):845-848.
- Tamblyn, K. C., and D. E. Conner. 1997. Bactericidal activity of organic acids against *Salmonella* Typhimurium attached to broiler chicken skin. *J. Food Prot.* 60(6):623-633.
- Tauxe, R. V. 1992. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. Pages 9-19 in *Campylobacter jejuni: current status and future trends*. I. Nachamkin, M. J. Blaser, and L. S. Tompkins, eds. American Society for Microbiology. Washington, D.C.
- Taylor, J. B., and D. W. Wohlers. 2004. Toxicological profile for chlorine dioxide and chlorite. U.S. Department of Health and Human Services.
<http://www.atsdr.cdc.gov/>.
- Thomas, C. J., and T. A. McMeekin. 1980. Contamination of broiler carcass skin during commercial processing procedures: an electron microscopic study. *Appl. Environ. Microbiol.* 40(1):133-144.
- Thomas, C. J., and T. A. McMeekin. 1982. Effect of water immersion on the microtopography of the skin of chicken carcasses. *J. Sci. Food Agr.* 33:549-554.
- Travers, K., and M. Barza. 2002. Morbidity of infections caused by antimicrobial resistant bacteria. *Clin. Infect. Dis.* 34(Suppl. 3):S131-S134.

United States Department of Agriculture (USDA). 1984. Regulations under the poultry products inspection act. Regulations Office, Policy and Program Planning, Food Safety and Inspection Service. U.S. Department of Agriculture. Washington, D.C.

United States Department of Agriculture (USDA). 2001. Retained water in raw meat and poultry products. Poultry chilling requirements. Final Rule. 9 CFR Parts 381 and 441. 66:1750-1772.

United States Department of Agriculture (USDA). 2002. The use of chlorine dioxide as an antimicrobial agent in poultry processing in the United States. USDA-FSIS, Office of International Affairs. November 2002.

United States Department of Agriculture (USDA). 2016a. Overview of the United States slaughter industry. <https://usda.mannlib.cornell.edu>.

United States Department of Agriculture (USDA). 2016b. Plain writing act compliance report 2016. <https://www.usda.gov/>.

United States Department of Agriculture (USDA). 2017a. Poultry production and value summary. National Agricultural Statistics Service. <https://mannlib.cornell.edu>.

United States Department of Agriculture (USDA). 2017b. Cost estimates of foodborne illnesses. <https://www.ers.usda.gov/data-products/cost-estimates-of-foodborne-illnesses/>.

United States Department of Agriculture (USDA). 2018. About the U.S. Department of Agriculture. <https://www.usda.gov/our-agency/about-usda>.

USDA-FSIS. 1996. Pathogen reduction: hazard analysis and critical control point (HACCP) systems, final rule. Fed. Regist. 61:38806.

USDA-FSIS. 2008. Compliance guidelines for controlling *Salmonella* and

- Campylobacter* in poultry. 2nd edition. <http://www.fsis.usda.gov/>.
- USDA-FSIS. 2014. Chicken from Farm to Table. U.S. Department of Agriculture, Food Safety and Inspection Service. <https://fsis.usda.gov/>.
- USDA-FSIS. 2015. Public health effects of raw chicken parts and comminuted chicken and turkey performance standards. <http://www.fsis.usda.gov/shared/PDF/Effects-Performance-Standards-Chicken-Parts-Comminuted.pdf>.
- USDA-FSIS. 2016a. Health hazard information sheet: peroxyacetic acid (PAA). FSIS Environmental, Safety and Health Group (ESHG). <http://www.fsis.usda.gov/>.
- USDA-FSIS. 2016b. Isolation and identification of *Campylobacter jejuni/coli/lari* from poultry rinse, sponge, and raw product samples. Laboratory Guidebook MLG 41.04.
- USDA-FSIS. 2018. Table of safe and suitable ingredients October 2018. https://www.fsis.usda.gov/wps/wcm/connect/ce40e7ae-3d55-419e-9c68-a1b6fefcd4de/7120.1_Table_2.pdf?MOD=AJPERES.
- Van de Giessen, A. W., B. P. Bloemberg, W. S. Ritmeester, and J. J. Tilburg. 1996. Epidemiological study on risk factors and risk reducing measures for *Campylobacter* infections in Dutch broiler flocks. *Epidemiol. Infect.* 117:245-250.
- Vandamme, P. 2000. Taxonomy of the family Campylobacteraceae. Pages 3-26 in *Campylobacter*. I. Nachamkin, and M. J. Blaser, eds. American Society for Microbiology. Washington, D.C.
- Vasseur, C., L. Baverel, M. Hebraud, and J. Labadie. 1999. Effect of osmotic, alkaline,

- acid, or thermal stress on the growth and inhibition of *Listeria monocytogenes*. J. Appl. Microbiol. 86:469-476.
- Veluz, G. A., S. Pitchiah, and C. Z. Alvarado. 2012. Attachment of *Salmonella* serovars and *Listeria monocytogenes* to stainless steel and plastic conveyor belts. Poultry Sci. 91(8):2004-2010.
- Vugia, D., A. Cronquist, J. Hadler, P. Blake, D. Blythe, K. Smith, D. Morse, P. Cieslak, T. Jones, D. Goldman, J. Guzewich, F. Angulo, P. Griffin, R. Tauxe, and K. Kretsinger. 2003. Preliminary FoodNet data on the incidence of infection with pathogens transmitted commonly through food - selected sites, United States. Morb. Mortal Wkly Rep. 53(16):338-343.
- Waldroup, A., C. Coleman, and K. Beers. 2000. Commercialization of cetylpyridinium chloride for use in the poultry industry. U.S. Food Additive Petition No. 2A4736, Appendix VII. Food and Drug Administration, Department of Health and Human Services. Washington, D.C.
- Wang, Y., C. Lehane, K. Ghebremeskel, and M. A. Crawford. 2010. Modern organic and broiler chickens sold for human consumption provide more energy from fat than protein. Public Health Nutr. 13(3):400-408.
- Wempe, J. M., C. A. Genigeorgis, T. B. Farver, and H. I. Yusufu. 1983. Prevalence of *Campylobacter jejuni* in two California chicken processing plants. Appl. Environ. Microbiol. 45(2):355-359.
- Whyte, P., J. D. Collins, K. McGill, and C. Monahan. 2007. Assessment of sodium dichloroisocyanurate in the control of microbiological cross-contamination in broiler carcass immersion chilling systems. J. Food Safety. 22(1):55-65.

- Wideman, N., M. Bailey, S. F. Bilgili, H. Thippareddi, L. Wang, C. Bratcher, M. Sanchez-Plata, and M. Singh. 2016. Evaluating best practices for *Campylobacter* and *Salmonella* reduction in poultry processing plants. *Poult. Sci.* 95(2):306-315.
- Williams, A., and O. A. Oyarzabal. 2012. Prevalence of *Campylobacter* spp. in skinless, boneless, retail broiler meat from 2005 through 2011 in Alabama, USA. *BMC Microbiol.* 12:184.
- Willis, W. L., and C. Murray. 1997. *Campylobacter jejuni* seasonal recovery observations of retail market broilers. *Poult. Sci.* 76(2):314-317.
- Zamora-Sanabria, R., and A. M. Alvarado. 2017. Preharvest *Salmonella* risk contamination and the control strategies. Intech: Open Science: Chapter 11.
- Zeitoun, A., F. Devlieghere, and J. Debevere. 1996. Increasing the shelf-life of poultry meat using acid decontamination and modified atmosphere packaging. Pages 215-225 in *Microbial Methods for the Meat Industry, Concerted Action CT94-1456*. M. H., Hinton, and C. Rowlinson, eds. University of Bristol Press. Bristol.
- Zhao, T., and M. P. Doyle. 2006. Reduction of *Campylobacter jejuni* on chicken wings by chemical treatments. *J. Food Prot.* 69(4):762-767.
- Zhao, C., B. Ge, J. D. Villena, R. Sudler, E. Yeh, S. Zhao, D. G. White, D. Wagner, and J. Meng. 2001. Prevalence of *Campylobacter* spp., *Escherichia coli*, and *Salmonella* serovars in retail chicken, turkey, pork, and beef from the greater Washington, D.C. Area. *Appl. Environ. Microbiol.* 67(12):5431-5436.

CHAPTER 3

LOW PH PROCESSING AID TO LOWER THE PRESENCE OF NATURALLY OCCURRING *CAMPYLOBACTER* ON WHOLE BROILER CARCASSES ¹

¹ Landrum, M.A., N.A. Cox, J.L. Wilson, D.E. Cosby, M.E. Berrang, G.R. Gamble, M.J. Da Costa, G.M. Pesti and A. Hinton, Jr. 2018. *Advanced Food and Nutritional Sciences*. 3:7-13. Reprinted here with permission of the publisher.

Abstract

Campylobacter is a serious foodborne pathogen for which the USDA-FSIS has recently established stricter performance standards than in the past. Processors must implement effective treatment plans to lower the microbial levels of *Campylobacter* to meet these regulations. This study evaluated the low pH processing aid, CMS PoultryHresh™, to reduce *Campylobacter* on carcasses. Three groups of 6 carcasses were collected prior to the chiller and were each individually placed into a 38 L container with either 20 L tap water (pH = 7.3) or 20 L of CMS PoultryHresh™ solution (pH = 1.4) with air agitation for 20 seconds. An untreated group was the control. After treatment, drained carcasses were placed into plastic bags, and rinsed by hand in 400 mL of buffered peptone for 60 seconds. Rinsates were cultured for *Campylobacter* by direct plating on Campy-Cefex agar and enrichment in Bolton's broth. If no *Campylobacter* were detected by direct plating, the incubated broth was plated onto Campy-Cefex agar and incubated under the same conditions. Confirmed *Campylobacter* were detected on 30/36 (83.3%) untreated carcasses, on 25/36 (69.4%) water treated carcasses, and on 2/36 (5.6%) CMS PoultryHresh™ treated carcasses. Treatment with PoultryHresh™ may be an option for processors to meet requirements and minimize *Campylobacter* presence to avoid regulatory action from FSIS.

Introduction

USDA-Food Safety and Inspection Service (FSIS) has established new performance standards for the presence of *Campylobacter* and *Salmonella* on fresh poultry. Processors have responded by continuing to develop and test intervention

methods that lower the prevalence of foodborne microorganisms in order to be in compliance with new regulations. *Campylobacter* is a major pathogen of concern which causes foodborne illness with gastrointestinal symptoms to more than 850,000 people in the U.S. each year (USDA, 2017). *Campylobacter* is most often reported being associated with poultry and has been found naturally occurring on as many as 88% of chicken carcasses (Guerrero-Legarreta, 2010). Through the past seven years, USDA-FSIS has encouraged greater control of food safety measures, improved record keeping, proper labeling requirements, correct testing methods, and higher performance standards (USDA, 2016). These regulations were made to reduce illnesses associated with foodborne pathogens found in poultry products and prevent an estimated 50,000 illnesses each year (USDA, 2016). More specifically, recent regulatory changes aim to achieve a 32% reduction of illnesses from *Campylobacter* alone (USDA, 2016). It is essential for broiler processing companies to establish an effective treatment plan to lower microbial levels of *Campylobacter* to pass rigorous performance standards set by USDA-FSIS. FSIS predicted that 46% of broiler companies will not be able to meet proposed 2016 performance standards for *Campylobacter* (Almanza, 2015).

Under regulations enacted in 2016, processing facilities are designated into categories 1, 2, 3, 4 or 5 by whether their microbial performance is consistent, variable, highly variable, passing, or failing to achieve FSIS *Campylobacter* standards (FSIS, 2016). Sampling to evaluate microbial control methods will become routine throughout the year, rather than infrequent sampling on consecutive days (USDA, 2016). Results will be reported to the public, heightening the importance of reducing microbial levels to reach set standards and avoiding consumer rejection. Being listed on the FSIS website as

“failing” to meet standards may have consequences for a company’s brand loyalty. The objective of this study was to evaluate a twenty second air agitated immersion in a proprietary antimicrobial treatment blend of sulfuric acid and copper sulfate, PoultrypHresh™, at a pH of 1.4 as a means to reduce or eliminate viable *Campylobacter* on whole broiler carcasses.

Materials and Methods

Treatment Procedures

Eighteen whole, pre-chill broiler carcasses were randomly collected from the evisceration line of a commercial processing facility prior to chilling, individually bagged, placed on ice and transported to the laboratory. Within 60 minutes, six whole carcasses were individually dipped for 20 seconds into 38 L, high density polyethylene plastic containers holding 20 L of tap water at a pH of approximately 7.3. Water was used as a treatment to determine the effects of rinsing. An additional 6 whole carcasses were dipped into similar containers for 20 seconds with 20 L of PoultrypHresh™ solution at a pH of 1.4. Air agitation of the solution in each bucket was achieved by pumping air (50 psi) from a compressor, through a six way manifold to the bottom of each container by means of plastic tubing (1/4” ID, McMaster-Carr, Elmherst, IL) which was secured to the side of the bucket by a metal pipe placed such that the tubing air exit was at the bottom of each container. After treatment (20 second immersion with agitation), each carcass was removed, and allowed to drip for 5 seconds before being subjected to a whole carcass rinse procedure in 400 mL of buffered peptone water (BPW) for 60 seconds using a mechanical rinsing machine (Dickens et al., 1985). The final six carcasses served as

untreated controls, receiving no dip treatment before whole carcass rinse sampling, as described above. Six replications were conducted on 6 separate days, n = 36 per treatment.

Campylobacter Detection Methods

Rinsates from each whole carcass were treated as described in the FSIS *Microbiology Laboratory Guidebook* (MLG, 2016). Initial rinses were used to spread 0.25 mL on four Campy-Cefex plates which were incubated microaerobically (5% O₂, 10% CO₂, 85% N₂) for 48 hours at 42°C. Thirty mL of each carcass rinsate was also placed into a tissue culture flask with 30 mL of 2X Bolton's broth (2X-BEB) and incubated concurrently with the direct plates under the same conditions. Incubated enrichment broth was plated onto a Cefex agar plate and incubated for 48 hours under the same conditions. After incubation, plates were examined for typical *Campylobacter* colonies, which were confirmed as members of the genus *Campylobacter* by observation of typical cellular morphology, motility under phase contrast microscopy and by use of a latex agglutination test kit (Microgen Bioproducts Ltd, Camberley, U.K.).

Statistical Analysis

The study was replicated six times using naturally contaminated *Campylobacter* carcasses, totaling 36 whole birds for each treatment (N = 108, n = 36). In each replicate, 6 untreated, 6 tap water treated, and 6 treated with PoultrypHresh™ at pH 1.4 were used. Each sample was recorded as either positive or negative for the presence of *Campylobacter* and data were analyzed using the Chi Square test for independence. Data

were analyzed using a generalized mixed model (binomial distribution) where treatment was considered as a fixed effect and the treatment nested within the days of collection as a random effect. The GLIMMIX Procedure of SAS (SAS Inst. Inc., Cary, N.C.) was used: Proc GLIMMIX Data=campy; Class day rep trt; Model status = trt /dist=bin link=logit solution; Random day (trt).

Results and Discussion

There was no difference in the presence of *Campylobacter* on the control or water treated samples from replication to replication. The covariance of treatment within replications was not significant ($p = 0.427$). Overall, data from all six replicates indicated that 30/36 (83.3%) of untreated carcasses, 25/36 (69.4%) water treated, and 2/36 (5.6%) treated with PoultryHresh™ were positive with *Campylobacter*. PoultryHresh™ met FSIS standards for *Campylobacter*, which have a maximum acceptance of 15.7% in broiler carcasses (Almanza, 2015). These data demonstrate that an agitated dip treatment of PoultryHresh™ significantly ($P < 0.001$) lowered the presence of *Campylobacter* on whole bird carcasses compared to either untreated or water treated carcasses (Table 3.1). Bird rinse samples directly plated with no enrichment demonstrated that 33.3% of untreated and 22.2% water treated carcasses were found to be positive for *Campylobacter*, while one of the 36 carcasses (2.8%) treated with PoultryHresh™ at pH 1.4 were positive. Once rinsates were enriched using 2X Bolton's broth, 75.0% of non-treated and 60.7% of water treated carcasses were contaminated with *Campylobacter*, while only 2.9% of carcasses treated with PoultryHresh™ were contaminated.

The present study demonstrated that PoultrypHresh™ could significantly ($P \leq 0.05$) reduce the presence of natural *Campylobacter* on broiler carcasses. These data show that an antimicrobial dip can be effective in the elimination of *Campylobacter* and may be an option for lowering the prevalence in commercial processing facilities. PoultrypHresh™ applied as an agitated dip treatment for whole broiler carcasses at pH 1.4 for 20 seconds can significantly ($P \leq 0.05$) lower the number of *Campylobacter* positive broiler carcass rinse samples. A single hurdle application treatment of this antimicrobial may reduce the presence of *Campylobacter* on whole carcasses, helping poultry processors to succeed in meeting regulatory requirements. Additional PoultrypHresh™ applications at multiple intervention points throughout processing plants may further increase microbial reductions.

References

- Almanza, A. V. 2015. Changes to the *Salmonella* and *Campylobacter* verification testing program. Food Safety and Inspection Service. 80(46):3940-3950.
- Dickens, J. A., N. A. Cox, J. S. Bailey, and J. E. Thomson. 1985. Automated microbiological sampling of broiler carcasses. Poult. Sci. 64(6):1116-1120.
- Food Safety and Inspection Service (FSIS). 2016. New performance standards for *Salmonella* and *Campylobacter* in not-ready-to-eat comminuted chicken and turkey products and raw chicken parts and changes to related agency verification procedures. United States Department of Agriculture. Fed. Reg. 81:7285-7300.

Guerrero-Legarreta, I. 2010. Handbook of poultry science and technology: secondary processing. John Wiley and Sons, Inc. 2:599-600.

MLG 41.04 United States Department of Agriculture. 2016. Laboratory Guidebook.

Isolation and identification of *Campylobacter jejuni/coli/lari* from poultry rinse, sponge, and raw product samples. <http://www.fsis.usda.gov/>.

National Institute of Allergy and Infectious Diseases (NIH). 2015. Campylobacteriosis.

United States Department of Health and Human Services. <https://www.niaid.nih.gov/topics/campylobacteriosis/Pages/default.aspx>.

United States Department of Agriculture (USDA). 2016. USDA finalizes new food safety measures to reduce *Salmonella* and *Campylobacter* in poultry.

<http://www.usda.gov/wps/portal/usda/>.

United States Department of Agriculture (USDA). 2017. Cost estimates of foodborne illnesses. <https://www.ers.usda.gov/data-products/cost-estimates-of-foodborne-illnesses/>.

Table 3.1. Number of direct, enriched, and overall samples positive for *Campylobacter* in untreated, water treated, and PoultrypHresh™ treated at pH 1.4 for 6 study replications. Samples not positive on direct analysis were enriched and re-evaluated.

Treatment	Direct		Enriched		Overall		Pr > t ¹
	+	-	+	-	+	-	
Control	12	24	18	6	30	6	-
Water	8	28	17	11	25	11	0.222
PoultrypHresh™	1	35	1	34	2	34	<0.001

¹ Probability that the treated groups are not different from the untreated control.

CHAPTER 4
TREATMENT WITH A LOW PH PROCESSING AID TO REDUCE
CAMPYLOBACTER COUNTS ON BROILER PARTS ²

² M.A. Landrum, N.A. Cox, D.E. Cosby, M.E. Berrang, and S.M. Russell. 2016. *Poultry Science*. 96(4):1028-1031. Reprinted here with permission of the publisher.

Abstract

New regulations and performance standards for *Campylobacter* have been implemented by the USDA - Food Safety and Inspection Service (FSIS). The objective of this study was to evaluate treatment with a low pH processing aid (CMS PoultryHresh™), a formulated low pH processing aid, to reduce numbers of *Campylobacter* on poultry carcasses, which could help companies meet regulatory requirements. Two experiments (3 replicates each) were conducted. Experiment 1, in each of 3 replicates, skin-on split chicken breasts (n = 15) were obtained from a local grocery and divided into groups of 5. The skin of each part was inoculated with approximately 10^7 cells of a gentamicin resistant *C. coli* (CC^{GR}) marker strain in an area of approximately 6.5 cm². CC^{GR} cells were allowed to attach for 5 minutes prior to treatment. Ten inoculated breasts were individually placed into separate 6 L plastic storage boxes containing either 3.5 L deionized water or PoultryHresh™ solution at a pH of 1.4. Parts were subjected to agitation (bubbled air) for 25 seconds. After treatment, each part was removed, allowed to drain for 5 seconds, and placed into a plastic bag prior to mechanical rinsing with 150 mL of BPW for 60 seconds. Five inoculated breasts served as controls, were untreated with a dip or agitation and sampled as above. Experiment 2 procedures were repeated using skin-on thighs under the same conditions. Rinsates were collected from each chicken part, serially diluted and plated onto Campy Cefex agar with 200 ppm gentamicin (CCGen). All plates were incubated microaerobically (5% O₂, 10% CO₂, 85% N₂) for 48 hours at 42°C, colonies were counted and the cfu/mL was log transformed. The use of PoultryHresh™ on split breast produced a 99.6% reduction compared to untreated controls, while thighs showed a

99.4% reduction in *Campylobacter* prevalence. This study demonstrated an approximate 2.5 log reduction ($P \leq 0.05$) using a 25 second air agitation treatment in PoultryHresh™ at pH 1.4 with no observable damage, which will help processors meet FSIS regulations.

Introduction

Although relatively sensitive to environmental stressors, *Campylobacter* is a food safety concern for consumers and one of the leading causes of bacterial foodborne diarrheal illnesses in the U.S. (Bashor et al., 2004; USDA, 2013). The USDA-FSIS is charged with reducing foodborne illnesses and thus has proposed new performance standards have been proposed for pathogen reduction in raw products, as 80% of poultry products distributed to consumers are sold in the raw form (Almanza, 2015). An assortment of chemicals are currently being used in poultry processing facilities (Peracetic acid, chlorine, Citrilow, Cecure, etc.).

The new regulations will require processing establishments to reassess Hazard Analysis and Critical Control Point (HACCP) plans for processes, which will improve reductions in pathogen numbers on final product (Almanza, 2015). Sampling will be conducted within individual processing establishments by FSIS inspection program personnel (IPP) on a routine basis (FSIS, 2015). Accepted performance standards specified by FSIS state the maximum percentage of poultry parts allowed to test *Campylobacter* positive is 7.7% (4 of 52 samples) (FSIS, 2015). FSIS estimates 46% of poultry establishments will be unable to meet the new standards and will be required to undergo production-processing changes to lower *Campylobacter* prevalence (Almanza, 2015). Such changes may include additional cleaning procedures, applying chemical

antimicrobials, and/or providing additional sanitation training to employees. If the facility fails to meet the standards, immediate follow-up sampling will be conducted by FSIS.

PoultryHresh™ is an antimicrobial blend of sulfuric acid and copper sulfate, liquid additive made from GRAS ingredients applied directly to food products to minimize or eliminate an assortment of foodborne pathogens. It is primarily used as a dip to regulate an acidic, low pH environment aimed to lower microorganism levels at intervention points during poultry processing. The purpose of this study was to evaluate whether *Campylobacter* levels on broiler parts may be lowered using the proprietary antimicrobial treatment, PoultryHresh™, as an immersion dip with air agitation at pH 1.4.

Materials and Methods

Bacterial Strain and Maintenance

A marker strain of *Campylobacter coli* (CC^{GR}), resistant to gentamicin (Cox et al., 2009) was used throughout this study. The culture was streaked onto Campy Cefex Agar (Stern et al., 1992) containing 200 ppm gentamicin (CCGen) (Sigma, St. Louis, MO) to restrict the growth of extraneous bacteria. Cultures were allowed to grow on plates at 42°C for 48 hours in a resealable plastic bag flushed with a microaerobic environment (5% O₂, 10% CO₂, 85% N₂), for use in the preparation of inocula for these experiments. Stock cultures were maintained at –80°C in *Campylobacter* Bolton's broth (Oxoid Ltd. Basingstoke, Hampshire, England) without supplements with 15% glycerol.

Inoculation of Parts

Skin-on split chicken breasts or thighs (n = 15) were obtained from a local grocery and divided into three groups of 5. Three replications were conducted on separate days. An area of approximately 6.5 cm² was denoted with a template on the skin's surface of each individual part and was inoculated using 0.1 mL of approximately 10⁷ cfu/mL CC^{GR} marker strain confirmed by serial dilution and plating onto CCGen plates, incubating at 42°C under microaerobic environment for 48 hours. CC^{GR} cells were allowed to stand for 5 minutes at 24°C to allow attachment to the skin prior to any treatment.

Treatment Procedures

Five inoculated split breast fillets or thighs were placed into separate 6 L plastic storage boxes, containing 3.5 L of deionized water each, five inoculated breast fillets or thighs were placed into similar individual storage containers containing 3.5 L of PoultryHresh™ solution at a pH of 1.4. A water dip was used to determine reductions associated with a rinsing effect. Diagonally across the bottom of each container was a Costar Stripette 5 mL pipette (Corning Corp., Corning, N.Y.) with 14 holes drilled along the length of the pipette (25 gauge needle), attached to an air compressor hose (approximately 125 psi). Air was pushed out of the holes causing agitation throughout each container for the 25 seconds of treatment. After treatment, each part was drained for 5 seconds, placed into a plastic freezer bag and rinsed with 150 mL of buffered peptone water for 60 seconds using a mechanical rinsing machine (Dickens et al., 1985). Five

breast fillets or thighs served as untreated controls and were untreated with any dip or agitation, but sampled as above. Three replications for both breasts and thighs were done.

Plating and Incubation

Rinsates were collected from each individual part, serially diluted and 0.1 mL plated onto CCGen agar plates. All plates were incubated microaerobically for 48 hours at 42°C. Characteristic CC^{GR} colonies were counted and the cfu/mL were log transformed. CC^{GR} was confirmed by observation of characteristic cellular morphology and motility under phase contrast microscopy.

Statistical Analysis

The study constructed three replicates of split breast and three replicates of thighs using 45 individual parts (N = 45, n = 5). Each replicate consisted of 5 untreated parts, 5 dipped in agitated water, and 5 treated with PoultrypHresh™ at pH 1.4. Duplicate counts were averaged and numbers were transformed by log₁₀. Data were analyzed using Statistica software (Statistica, 2013). A General Linear Model was conducted to determine whether methods were significantly different ($P \leq 0.05$). Means were separated with a Duncan multiple range test; statistical significance was assigned at $P \leq 0.05$.

Results and Discussion

The current study showed that PoultrypHresh™ antimicrobial could be used to significantly ($P \leq 0.05$) reduce the numbers of CC^{GR} recovered from parts. The two parts

used in this study, breasts and thighs, reflect future FSIS testing, which will focus on high production parts including breasts and thighs.

PoultryHresh™ on split breasts reduced the level of *C. coli* by 99.6% reduction when compared to CC^{GR} levels recovered from untreated breasts (Table 4.1).

PoultryHresh™ was also significantly ($P \leq 0.05$) more effective than the water wash controls, showing a 97.9% reduction in the three replications. Therefore, a

PoultryHresh™ dip resulted in an average 2.41 log₁₀ cfu/mL reduction of *C. coli* compared to untreated split breasts and 1.72 log₁₀ cfu/mL reduction compared to water treatment (Table 4.1).

Similar results were noted when testing skin-on thighs; treating with PoultryHresh™ significantly lowered CC^{GR} levels. When comparing thighs treated with PoultryHresh™ to untreated thighs, the reduction of CC^{GR} colonies recovered was 99.3%. There was a 95.5% reduction in thighs dipped into PoultryHresh™ at pH 1.4 compared to thighs treated with a water dip. Overall, there was a 2.16 log₁₀ cfu/mL reduction in CC^{GR} colonies of untreated thighs and 1.41 log₁₀ cfu/mL reduction from water treated thighs with air agitation when compared to a dip in the PoultryHresh™ product (Table 4.1).

There are several factors that contribute to the number of *Campylobacter* on poultry products in processing plants, including, but not limited to sanitation practices and antimicrobial use (Marriott, 2013). Reports show processors consistently produce *Campylobacter* positive raw products, although FSIS has encouraged companies to reduce the level of *Campylobacter* in slaughter operations (Bennett, 2008). Once the new FSIS performance standards are fully implemented, companies that fail to meet standards

must make changes in the process that lower *Campylobacter* prevalence (Almanza, 2015). FSIS reports suggest that the most likely corrective action will be the addition of antimicrobials within the processing facility (Almanza, 2015).

Water agitation and pH adjustments are factors shown to improve microbial reductions, specifically targeting *Campylobacter*, throughout secondary processing (Bennett, 2008). Studies comparing the efficacy of spray versus dip applications concluded that there were significantly ($P \leq 0.05$) greater bacterial reductions when using immersion treatments (Singh, 2013). Multiple hurdles heighten the reduction or elimination of *Campylobacter* counts, rather than a single control measure. Air agitated PoultrypHresh™ employs several hurdles to reduce microbial prevalence with no adverse change of appearance including antimicrobial properties, pH control, brief exposure time, and air agitation. The method tested here of compressed air bubbled through an immersion vessel containing PoultrypHresh™ resulted in approximately a 2.5 log₁₀ cfu/mL reduction of CC^{GR} counts for breasts and thighs compared to untreated inoculated parts. These reductions should be sufficient to allow most processors to meet the new FSIS standards of pathogen reduction. PoultrypHresh™ was used to test parts for *Salmonella*, but percent reductions were not as dramatic as shown in *Campylobacter*. Future research on a larger scale in a commercial processing plant is planned to further assess the viability of this low-acid processing aid to reduce *Campylobacter* on poultry parts and/or carcasses. PoultrypHresh™ could be part of a successful program to reduce microbial contamination on cut-up broiler parts.

References

- Almanza, A. 2015. Changes to the *Salmonella* and *Campylobacter* verification testing program. Food Safety and Inspection Service, USDA. 80:3940-3950.
- Bashor, M. P., P. A. Curtis, K. M. Keener, B. W. Sheldon, S. Kathariou, and J. A. Osborne. 2004. Effects of carcass washers on *Campylobacter* contamination in large broiler processing plants. *Poult. Sci.* 83:1232-1239.
- Bennett, P. 2008. Compliance guidelines for controlling *Salmonella* and *Campylobacter* in poultry. 2nd ed. http://www.fsis.usda.gov/wps/wcm/connect/1d562776-ebfb4ea5-a19b-e994776a02c6/Compliance_Guideline_Controlling_Salmonella_Poultry.pdf.
- Cox, N. A., L. J. Richardson, M. E. Berrang, P. J. Fedarka-Cray, and R. J. Buhr. 2009. *Campylobacter coli* naturally resistant to elevated levels of gentamicin as a marker strain in poultry research. *J. Food Prot.* 72:1288-1292.
- Dickens, J. A., N. A. Cox, J. S. Bailey, and J. E. Thomson. 1985. Automated microbiological sampling of broiler carcasses. *Poult. Sci.* 64:1116-1120.
- Food Safety Education - *Campylobacter* question and answers. 2013. United States Department of Agriculture (USDA). <http://www.fsis.usda.gov/>.
- Food Safety and Inspection Service (FSIS). 2015. Pathogen Reduction - *Salmonella* and *Campylobacter* performance standards verification testing. United States Department of Agriculture.
- Marriott, N. 2013. Principles of Food Sanitation. 3rd ed. Springer Science & Business Media.

Singh, M. 2013. Intervention strategies for control of *Campylobacter* during poultry processing. Department of Poultry Science. Auburn University. https://confexrecordings.s3.amazonaws.com/asas/RM13/paper471_1.pdf .

Statistica. 2013. StatSoft, Inc. Version 12.0, ed. Tulsa, OK, USA.

Stern, N. J., B. Wojton and K. Kwiatek. 1992. A differential selective medium and dry ice generated atmosphere for recovery of *Campylobacter jejuni*. J. Food Prot. 55:514-517.

Table 4.1. Average Log₁₀ cfu/mL of *Campylobacter coli* recovered from parts dip treated with no treatment, water, or PoultrypHresh™ for 25 seconds with agitation in 3 study replications (mean ± standard error). ^{a, b, c, x, y, z} values with different subscripts are significantly ($P \leq 0.05$) different by Duncan's Multiple Range test in parts.

Part	Treatment	Average Log ₁₀ (cfu/mL)	Reduction from Untreated (%)	Reduction from Water (%)
Split Breast	Untreated	5.09 ^a ± 0.18	-	-
	Water	4.39 ^b ± 0.18	80.0	-
	PpH ¹	2.67 ^c ± 0.13	99.6	98.1
Thigh	Untreated	4.70 ^x ± 0.09	-	-
	Water	3.95 ^y ± 0.06	82.2	-
	PpH	2.54 ^z ± 0.24	99.3	96.1

¹ PpH = CMS PoultrypHresh™

CHAPTER 5
REDUCTION OF *CAMPYLOBACTER* PREVELANCE ON CHICKEN LIVERS USING
A LOW ACID PROCESSING AID ³

³ Landrum, M.A., N.A. Cox, D.E. Cosby, M.E. Berrang, A. Hinton Jr., S.C. Mize and J.S. Jackson. 2018.
Advanced Food and Nutritional Sciences. 3:1-6. Reprinted here with permission of the publisher.

Abstract

Chicken livers have become a prime source for *Campylobacter* outbreaks and products are needed to allow processors a more efficient way of controlling foodborne pathogens associated with livers. *Campylobacter* prevalence reductions on livers treated with a low pH processing aid (CMS PoultrypHresh™), with and without a surfactant (PoultrypHresh™ Plus) were studied. Chicken livers (n = 13/treatment group) were individually inoculated with a *C. coli* marker strain (10^7) and dipped into sterile cups containing 100 mL of water, PoultrypHresh™ or PoultrypHresh™ Plus for 15 seconds, removed and allowed 5 seconds to drain. Each liver was placed into 50 mL buffered peptone water and hand shaken for 60 seconds; controls (n = 10) same procedure, no treatment. Rinsates were serially diluted and plated onto Campy Cefex agar with 200 ppm gentamicin. Plates were incubated for 48 hours at 42°C microaerobically, colonies counted and log transformed. Procedures were replicated 3 times. Significant reductions in the prevalence of *C. coli* in livers treated with PoultrypHresh™ and PoultrypHresh™ Plus compared to untreated was 98.1% and 99.4%, respectively and with no change in appearance. Treating with PoultrypHresh™ may allow processors to meet rising performance standards on poultry livers.

Introduction

Campylobacter, the third leading cause of foodborne illness in the U.S., continues to be a major concern to the poultry industry. Illnesses and/or outbreaks often occur from consumption of raw or undercooked products (Little et al., 2010; Simaluiza et al., 2015) and may cause symptoms including diarrhea, abdominal cramping, fever and vomiting

(Noormohamed and Fakur, 2012). Poultry products are commonly implicated with outbreaks of campylobacteriosis and investigations worldwide have demonstrated how chicken liver is increasingly becoming a prime source for contamination (Little et al., 2010; Firlieyanti et al., 2016). In the U.S., *Campylobacter* was found prevalent in 77% of livers (Harrison et al., 2013), while another study reported *Campylobacter* in as many as 92.9% of commercial chicken livers (Fernández and Pisón, 1996). The U.K. experienced a substantial increase in campylobacteriosis associated with liver dishes between 2009 and 2011, causing the Food Standards Agency (FSA) to categorize liver as a high-risk food product (Edwards et al., 2014). The Centers for Disease Control and Prevention (CDC) investigated and reported outbreaks of *Campylobacter* infections from poultry liver in the U.K. and Australia (CDC, 2013). In Switzerland, *Campylobacter* was isolated in livers from 10% to 100%, varying by season (Harrison et al., 2013). Liver is also a prime source for *Campylobacter* outbreaks in England (Noormohamed and Fakur, 2012). Therefore, microbial contamination of broiler livers is a worldwide concern for the industry.

The presence of *Campylobacter* on chicken liver has become a widespread problem and a public health concern. Often cases of campylobacteriosis go undetected and are not reported; therefore illness and outbreak numbers are probably even higher than predicted, further increasing the problems caused by this foodborne pathogen (Bryan and Doyle, 1995). Proper procedures for eliminating *Campylobacter* from livers is thoroughly cooking until an internal temperature exceeding 70°C is reached for a minimum of 2 minutes (Whyte et al., 2006; Harrison et al., 2013; Hutchison et al., 2015). One factor aiding the problem is many liver recipes recommend cooking by ‘flash

frying', which allows livers to maintain a pink internal color (Strachan et al., 2012; Harrison et al., 2013; Hutchison et al., 2015). This cooking method is not adequate to eliminate *Campylobacter*, allowing it to infect the consumer (Whyte et al., 2006). This assists in explaining the large number of campylobacteriosis outbreaks in individuals who recently attended catered events or consumed restaurant meals. Research has found that caterers or restaurant cooks are likely to undercook livers in an attempt to maintain the pink coloration consumers desire (Hutchison et al., 2015). This, however, allows many individuals to become sick or an outbreak to occur. Hutchinson et al. (2015) described a variety of essential oils and antimicrobial ingredients evaluated as additives for liver recipes, although significant reductions were not found.

There are some intervention strategies, which include freezing, alternative cooking methods, boiling, chlorinated water, organic acid treatment or pre-cooking treatments (Bryan and Doyle, 1995; Noormohamed and Fakur, 2012; Harrison et al., 2013; Hutchison et al., 2015; Firlieyanti et al., 2016). Further research demonstrated *Campylobacter* to be more prevalent on a liver's outer surface than internally (Thompson et al., 2018). This indicates external treatment methods may exhibit greater reductions. Past research evaluating organic acid treatment on livers exhibited changes in the surface coloration post treatment. This surface lightening was described as "bleaching" (Hutchison et al., 2015). This study investigates the reduction of *Campylobacter* on livers treated with a low acid processing aid, CMS PoultryHresh™, and the product containing the addition of a surfactant (Glucopon), PoultryHresh™ Plus. PoultryHresh is made from GRAS ingredients, such as sulfuric acid and copper sulfate, applied directly to food products to reduce foodborne pathogens. Differences in past research and this study

include the acid used and the length of immersion. Past research used a duration dip time of 2 minutes, whereas the current research is only 15 seconds (Hutchison et al., 2015). If effective, the treatment could potentially prevent cross-contamination in a consumer kitchen and lower *Campylobacter* prevalence internally, as it is not conclusive whether outer contamination seeps into the liver. This treatment may assist processors in reducing contamination levels within processing facilities, as dip time is rapid and potentially a reasonable addition for processing procedures.

Materials and Methods

Bacterial Strain

The bacterial *Campylobacter* strain used for this research is a gentamicin resistant marker strain, *Campylobacter coli* (CC^{GR}), obtained from Dr. Nelson Cox, USDA, Athens, GA (Cox et al., 2009). Initially, CC^{GR} was streaked onto Campy Cefex Agar (Stern et al., 1992) containing 200 ppm gentamicin (CCGen) (Sigma, St. Louis, MO). The culture was incubated microaerobically for 48 hours at 42°C (5% O₂, 10% CO₂, 85% N₂). Forty-eight hour (\pm 4 hours) plates of this culture were used to prepare the inocula for this research.

Inoculation of Parts

Livers were obtained from a local grocery (N = 49), livers were divided into 4 groups; Treatment 1 – CMS PoultrypHresh™ (PpH) (n = 13); Treatment 2 – PpH w/surfactant (PpH Plus) (n = 13) and Treatment 3 – inoculated untreated control (n = 10). The surfactant added to produce PoultrypHresh™ Plus was glucoPON at 0.2%. A 10^8

suspension of CC^{GR} (0.1 mL) was used to individually inoculate the surface of each liver. Livers were left undisturbed for 5 minutes to allow the cells an adequate attachment period.

Treatment

Thirteen livers were placed into separate specimen cups containing 100 mL of either water, PpH or PpH Plus for 15 seconds with no agitation. Water was used as a treatment to determine the effects of rinsing. When removed, livers were allowed to drain for 5 seconds and placed into individual sterile specimen cups containing 50 mL of buffered peptone water. Each liver was hand shaken for 60 seconds. The controls were inoculated the same as the experimental groups but were not subjected to any treatment before being placed into the specimen cups for rinsing.

Plating and Incubation

After hand rinsing, each rinsate was collected, serially diluted and plated onto Campy Cefex agar with 200 ppm gentamicin. Plates were incubated microaerobically at 42°C for 48 hours. Colonies were counted and cfu/mL data was log transformed. All procedures were replicated 3 times.

Results

Since *Campylobacter* in chicken livers is quickly becoming a major concern in the food industry, this study evaluated a potential treatment option to lower prevalence and chance of infection. Results showed the average recovery of *C. coli* on livers

receiving no treatment was 5.5 log₁₀ cfu/mL (Table 5.1). After livers were treated with a PpH 15 second dip, the recovery was reduced to 3.9 log₁₀ cfu/mL. Livers that received a PpH Plus 15 second dip were found to have *Campylobacter* recovery levels lowered to 3.3 log₁₀ cfu/mL. A large amount of foaming was noted when surfactant was added to PpH at this concentration, which was not ideal for a processing facility. These results indicate a 1.7 log₁₀ cfu/mL reduction (98.1%) when using a PpH dip and a 2.2 log₁₀ cfu/mL reduction (99.4%) dipping with PpH Plus compared to untreated samples. When treated results were compared with a 15 second water dip, PpH reduced the average log₁₀ cfu/mL by 91.9% (1.1 log₁₀ cfu/mL), while PpH Plus lowered *C. coli* by 97.5% (1.6 log₁₀ cfu/mL). No visible organoleptic damage was demonstrated or reported post-treatment. Table 5.1 shows average recovery of *C. coli* from all replicates averaged.

Discussion

A treatment capable of providing substantial reductions in *Campylobacter* levels is important for poultry producers and consumers worldwide. Alternate treatment options may include the use of chlorinated water, although Bryan and Doyle (1995) found that while chlorine may assist in lowering cross-contamination between carcasses, it has little effect on bacteria attached to the skin and muscle surfaces. Harrison et al. (2013) reported the method of freezing does reduce the presence of *Campylobacter* on the skin and muscle of the broiler. It is likely the consumer may prefer what is considered to be a fresh, never frozen product. Such findings, however, contradict those of Fernandez and Pison (1996) who found *Campylobacter* to be highly prevalent in frozen poultry liver.

Additional treatment options include the use of organic acid, which Firlieyanti et al. (2016) reported causes color changes and bleaching on the liver surface. Several studies showed it is not effective for internal *Campylobacter* reduction (Hutchison et al., 2015; Firlieyanti et al., 2016). Noormohamed and Fakhr (2012) discussed how foodborne pathogen resistance to antimicrobials is alarming and may arise from cross-contamination during processing, possibly causing consequences on human health. Their study also demonstrated the majority of *Campylobacter* isolates were resistant to five of the seven antimicrobials researched and 81 isolates were resistant to more than seven antimicrobials. This further increases the need for a treatment to effectively lower and/or eliminate *Campylobacter* before products are shipped from processing facilities, as Vashin et al. (2009) discussed how the likelihood of transferring *Campylobacter* rises in further stages of secondary processing.

Researchers have demonstrated the need to reduce prevalence of *Campylobacter* on poultry liver surfaces throughout the world. Illness and outbreaks arising from livers are increasingly becoming more prevalent and research has found the majority of retail livers are contaminated with *Campylobacter* at varying levels (Firlieyanti et al., 2016). While the key to *Campylobacter* elimination is allowing adequate cooking times and temperatures, recipes continue suggesting undercooking or flash cooking which could result in illnesses. Intervention strategies within the processing facility are important to lowering *Campylobacter* prevalence and the ample reductions demonstrated in this study may potentially provide the industry with an effective means to reduce the presence of this pathogen and hence human illness. Future research will further evaluate PpH on a larger scale to reduce *Campylobacter* contamination of poultry products. PpH may

provide another hurdle *Campylobacter* must cross in order to infect consumers, effectively reducing the number of campylobacteriosis illnesses associated with poultry products.

References

- Bryan, F. L., and M. P. Doyle. 1995. Health risks and consequences of *Salmonella* and *Campylobacter jejuni* in raw poultry. *J. Food Prot.* 58:326-344.
- Centers for Disease Control and Prevention (CDC). 2013. Multistate outbreak of *Campylobacter jejuni* infections associated with undercooked chicken livers-northeastern United States 2012. *Morb. Mortal. Wkly Rep.* 62:874-876.
- Cox, N. A., L. J. Richardson, M. E. Berrang, P. J. Fedorka-Cray, and R. J. Buhr. 2009. *Campylobacter coli* naturally resistant to elevated levels of gentamicin as a marker strain in poultry research. *J. Food Prot.* 72:1288-1292.
- Edwards, E. S., L. M. Milne, K. Morrow, P. Sheridan, N. Q. Verlander, R. Mulla, J. F. Richardson, A. Pender, M. Lilley, and M. Reacher. 2014. Campylobacteriosis outbreak associated with consumption of undercooked chicken liver pate in the East of England, September 2011: identification of a dose-response risk. *Epidemiol. Infect.* 142:352-357.
- Fernández, H., and V. Pisón. 1996. Isolation of thermotolerant species of *Campylobacter* from commercial chicken livers. *Int. J. Food Microbiol.* 29:75-80.
- Firleyanti, A. S., P. L. Connerton, and I. F. Connerton. 2016. Campylobacters and their bacteriophages from chicken liver: The prospect for phage biocontrol. *Int. J. Food Microbiol.* 237:121-127.

- Harrison, D., J. E. L. Corry, M. A. Tchórzewska, V. K. Morris, and M. L. Hutchison. 2013. Freezing as an intervention to reduce the numbers of campylobacters isolated from chicken livers. *J. Appl. Microbiol.* 57:206-213.
- Hutchison, M., D. Harrison, I. Richardson, and M. Tchórzewska. 2015. A method for the preparation of chicken liver pâté that reliably destroys *Campylobacters*. *Int. J. Environ. Res. Public Health.* 12:4652-4669.
- Little, C. L., F. J. Gormley, N. Rawal, and J. F. Richardson. 2010. A recipe for disaster: outbreaks of campylobacteriosis associated with poultry liver pâté in England and Wales. *Epidemiol. Infect.* 138:1691-1694.
- Noormohamed, A., and M. K. Fakur. 2012. Incidence and antimicrobial resistance profiling of *Campylobacter* in retail chicken livers and gizzards. *Foodborne Pathog. Dis.* 9:617-624.
- Simaluiza, R. J., Z. Toledo, S. Ochoa, and H. Fernandez. 2015. The prevalence and antimicrobial resistance of *Campylobacter jejuni* and *Campylobacter coli* in chicken livers used for human consumption in Ecuador. *J. Anim. Vet. Adv.* 14:6-9.
- Stern, N. J., B. Wojton, and K. Kwiatek. 1992. A differential selective medium and dry ice generated atmosphere for recovery of *Campylobacter jejuni*. *J. Food Prot.* 72:1288-1292.
- Strachan, N. J. C., M. MacRae, A. Thomson, O. Rotariu, I. D. Ogden, and K. J. Forbes. 2012. Source attribution, prevalence and enumeration of *Campylobacter* spp. from retail liver. *Int. J. Food Microbiol.* 153:234-236.
- Thompson, T. M., M. E. Berrang, N. A. Cox, and R. J. Meinersmann. 2018.

Campylobacter prevalence in retail chicken liver. Poult. Sci. (Suppl 1).

Vashin, I., T. Stoyanchev, C. Ring, and V. Atanassova. 2009. Prevalence of *Campylobacter* spp. in frozen poultry giblets at Bulgarian retail markets. Trakia J. Sci. 7:55-57.

Whyte, R., J. A. Hudson, and C. Graham. 2006. *Campylobacter* in chicken livers and their destruction by pan frying. Lett. Appl. Microbiol. 43:591-595.

Table 5.1. Average Log₁₀ cfu/mL of *Campylobacter coli* recovered from livers dip treated with no treatment, water, PoultrypHresh™, or PoultrypHresh™ Plus for 15 seconds with no agitation (mean ± standard error) in 3 study replications. ^{a, b, c, d} values with different subscripts are significantly ($P \leq 0.05$) different by Tukey's Honest Significant Difference test in livers.

Treatment	Average Log ₁₀ (cfu/mL)	Reduction from Untreated (%)	Reduction from Water (%)
Untreated	5.5 ^a ± 0.1	-	-
Water	4.9 ^b ± 0.1	76.0	-
PpH ¹	3.8 ^c ± 0.1	98.1	91.9
PpH Plus ²	3.3 ^d ± 0.2	99.4	97.5

¹ PpH = CMS PoultrypHresh™

² PpH Plus = CMS PoultrypHresh™ with surfactant

CHAPTER 6

REDUCTION OF *CAMPYLOBACTER* LEVELS ON POULTRY THIGHS USING SEQUENTIAL TREATMENTS OF ANTIMICROBIALS ⁴

⁴ Landrum, M.A., N.A. Cox, J.L. Wilson, M.E. Berrang, G.R. Gamble, M.A. Harrison, B.D. Fairchild, W.O. Kim, and A. Hinton, Jr. To be submitted to *Advanced Food and Nutritional Sciences*.

Abstract

Campylobacter is a major concern for poultry processors, as USDA performance standards have become more strict, currently allowing only 7.7% positive in poultry parts. CMS PoultryHresh™, a low pH processing aid, was evaluated in this study using consecutive and sequential dip treatments with peracetic acid to reduce *Campylobacter* in poultry thighs. Skin-on thighs (n = 3/treatment group) were inoculated individually with a *Campylobacter coli* marker strain (10⁸) and dipped into bags containing 1 L of the first treatment for 6 seconds. Thighs were allowed 5 seconds drip time and placed onto aluminum foil for 1 minute. Thighs were then dipped into the second treatment for 6 seconds, given 5 seconds drip time and placed into a bag with 150 mL buffered peptone water to be hand shaken for 60 seconds. Untreated thighs followed the same procedure, with no treatment. Rinsates were serially diluted, plated onto Campy Cefex agar with 200 ppm gentamicin and incubated microaerobically for 48 hours at 42°C. Procedures were replicated 5 times. Consecutive dips of PoultryHresh™ and PAA demonstrated reductions 98.2% and 99.3%, respectively, compared to untreated thighs. Treating with PoultryHresh™ followed by peracetic acid reduced *Campylobacter* by 99.2% from untreated thighs. Interestingly, dipping in the opposite sequence (peracetic acid then PoultryHresh™) significantly reduced *Campylobacter* more than any other treatment (99.9% from untreated). Results of this study showed sequential dipping in different chemicals could lead to increasingly greater reductions, findings not shown in previous research. PoultryHresh™ could be beneficial for processors, especially in a sequence dip followed by PAA for meeting performance standards.

Introduction

Campylobacter is the main cause of bacterial gastroenteritis in the world and researchers have shown it to be present in high levels in retail poultry (Harrison et al., 2001; Moore et al., 2002). Of *Campylobacter* illnesses, 50 - 70% were caused by consumption of poultry or poultry products (Allos, 2001; Humphrey et al., 2007). Symptoms of *Campylobacter* include fever, abdominal pain and diarrhea within 2 - 5 days of ingesting contaminated product (Robinson, 1981; Black et al., 1988). Stricter performance standards have been implemented by the USDA, aimed at reducing incidence of *Campylobacter* in processing facilities (FSIS, 2015). FSIS estimated 46% of poultry facilities would not be able to meet new requirements; therefore, processors must find efficient antimicrobial treatments to be used throughout the plant. It is mandatory that *Campylobacter* prevalence remain below 7.7% on poultry parts (4 of 52 samples), allowing only a small margin of error (Inspection Methods, 2018).

A variety of antimicrobials have been evaluated for use on poultry at various points throughout processing procedures. It is essential chemicals be cost effective, reduce pathogen prevalence and not cause organoleptic damage to poultry carcasses or parts (Bauermeister et al., 2008). Antimicrobials vary in levels of efficacy, treatment concentration, contact time and application method. Chemical efficacy is affected by microbial load, composition of flora, organic material or possibly changed from residual effects of prior treatments (Gill and Badoui, 2004). Stopforth et al. (2007) determined a single intervention could not significantly reduce pathogen presence on finished carcasses. Instead, a multi-hurdle approach is required at various intervention points throughout processing for increased pathogen reduction on poultry products (Bacon et al.,

2000; Stopforth et al., 2007). There was no previous research found evaluating consecutive, sequential treatments and how they affect pathogens. The purpose of this study was to evaluate the effectiveness of a low acid antimicrobial blend of sulfuric acid and copper sulfate, CMS PoultrypHresh™, and whether dipping carcasses in consecutive or sequential treatments with peracetic acid (PAA) leads to further reductions in the prevalence of *Campylobacter*.

Materials and Methods

Bacterial Marker Strain

Cox et al. (2009) developed a marker strain *Campylobacter* resistant to the antibiotic gentamicin, *Campylobacter coli* (CC^{GR}), used in this study. Campy Cefex agar (Sigma, St. Louis, MO) plates were prepared using 200 ppm gentamicin (CCGen) to eliminate the growth of background microflora which is naturally present on poultry thighs. Initial cultures were streaked from storage in an -80°C freezer, maintained in Bolton's broth with 15% glycerol and no supplements. Cultures were streaked onto CCGen agar and incubated in sealed bags under microaerobic (5% O₂, 10% CO₂, 85% N₂) conditions at 42°C for 48 hours. A sterile swab was used to remove colonies and mix them into a 9 mL tube of Bolton's broth, placed in sealed bags and incubated microaerobically for 24 hours at 42°C. Tubes were evaluated for colorimeter analysis by a Spectronic 200E (Thermo Fisher Scientific, Madison, WI) and approximately a 10⁸ cfu/mL CC^{GR} inoculum prepared. Inoculum was confirmed using serial dilutions, plated on CCGen and incubated for 48 hours at 42°C.

Treatment Procedures

Skin-on poultry thighs were purchased from a local grocery (n = 18) and divided into the six treatment groups. A 6.5 cm² section of skin was inoculated on each thigh with 0.1 mL of 10⁸ CC^{GR} marker strain. Each thigh was inoculated individually and given a 5 minute attachment period before treatment. Thighs were each dipped into individual Ziploc bags containing 1 liter of water, PAA (600 ppm), or PoultrypHresh (pH 1.5) for 6 seconds. Thighs were removed and placed on tin foil squares for 1 minute. Each thigh was then dipped into the second dip treatment for 6 seconds, drained 5 second, placed into individual sealable bags with 150 mL buffered peptone water (BPW), and hand shaken for 60 seconds. Thighs were removed, and rinses placed on ice for approximately 15 minutes before diluting. Three thighs served as controls in each replication and were inoculated as described, remained untreated and placed directly into rinse bags with BPW after the attachment period. Water was used as a treatment to determine the effects of rinsing. Procedures were replicated 5 times.

Plating and Incubation

Rinsates were serially diluted and 0.1 mL spread onto CCGen agar plates in duplicate. Plates were incubated at 42°C for 48 hours. Characteristic colonies were counted, and cfu/mL log transformed. Phase contrast microscopy was used to confirm any suspicious or uncharacteristic colonies, evaluating morphology and motility.

Statistical Analysis

The study was constructed of five replicates of skin-on thighs using 18 individual thighs (N = 15; n = 3). Each replicate consisted of 3 of each untreated, consecutive water dips, consecutive PoultrypHresh™ (pH 1.5) dips, consecutive PAA (600 ppm) dips, PoultrypHresh™ followed by PAA dip, and PAA followed by PoultrypHresh™ dip. Duplicate counts were averaged, and numbers were transformed by log₁₀. Data was analyzed using Statistica software (Statistica, 2013). A General Linear Model was conducted to determine whether sequential dips were statistically different ($P \leq 0.05$). Means were separated with a Tukey Multiple Comparison test; statistical significance was assigned at $P \leq 0.05$.

Results

Consecutive water dips were used to demonstrate whether water alone could reduce *Campylobacter* prevalence, finding a 0.8 log₁₀ cfu/mL reduction, although contamination remained high at 4.9 log₁₀ cfu/mL (Table 6.1). Treating with consecutive PoultrypHresh™ dips significantly ($P \leq 0.05$) reduced *Campylobacter* from 5.64 log₁₀ cfu/mL untreated and 4.87 log₁₀ cfu/mL water dipped to 3.90 log₁₀ cfu/mL (Table 6.1). This equates to a reduction of 98.2% from untreated and 89.3% from water dipped thighs (Table 6.1). Dipping with consecutive PAA dips showed slightly higher reductions; reducing *Campylobacter* levels by 99.3% (2.1 log₁₀ cfu/mL) from untreated and 95.7% (1.4 log₁₀ cfu/mL) from water dipped samples (Table 6.1).

Sequential dips using PAA and PoultrypHresh™ demonstrated very interesting results. Dipping in PoultrypHresh™ followed by PAA demonstrated findings similar to

consecutive PAA dips. However, when thighs were dipped in PAA followed by PoultrypHresh™, reductions were significantly ($P \leq 0.05$) higher than any other dipping combination. *Campylobacter* levels were reduced 99.9% ($> 3 \log_{10}$ cfu/mL reduction) from untreated thighs and 99.6% (2.4 \log_{10} cfu/mL reduction) from water treated (Table 6.1). This pattern was found in all 5 replications of the study, therefore the order of chemicals used in dipping sequences was significantly ($P \leq 0.05$) correlated to *Campylobacter* reductions.

Discussion

Results can be compared to those of Landrum et al. (2018), who demonstrated greater *Campylobacter* reductions compared to untreated thighs of 2.2 \log_{10} cfu/mL compared to 1.7 \log_{10} cfu/mL in this study. Comparing thighs treated with PoultrypHresh™ to water dipped demonstrated a 1.4 \log_{10} cfu/mL reduction by Landrum et al. (2018) compared to the 1.0 \log_{10} cfu/mL in this study. One difference in the two studies was the dip time (25 seconds in Landrum et al. (2018) compared to two, 6 second dips). Previous research also used bubbled air agitation, which was not used in this study. Shorten dip times of 6 seconds were chosen for this research due to the higher practicality of being used in a modern processing facility. The 25 second dip demonstrated better results, seemingly due to the increased exposure time, although the possibility remains that air agitation added to the significantly higher reductions. Previous researchers have demonstrated that water agitation improves microbial reductions during secondary processing procedures (Bennett, 2008). Perhaps, air agitation may heighten the reductions found in this study, a concept that could be evaluated in future research.

PAA is a low pH organic peroxide mixture of acetic acid and hydrogen peroxide currently being used throughout the U.S. in multiple processing facilities (Bauermeister et al., 2008; USDA-FSIS, 2016; Sukted et al., 2017). The mode of action for PAA is causing disruption to the cell membrane permeability, altering protein synthesis, leading to death (Oyarzabal, 2005). PAA reduced the prevalence of *Campylobacter* more than consecutive PoultryHresh™ dips, although differences were not significant. Findings by Bauermeister et al. (2008) showed a 1.5 log₁₀ cfu/mL *Campylobacter* reduction using an extremely low concentration of only 0.02% PAA. As PAA is approved for use throughout processing at concentrations up to 2,000 ppm, reductions could be even greater with increased acid levels (FSIS Directive, 2018). Bauermeister (2015) used 200 ppm PAA, only demonstrating a 1.5 log₁₀ cfu/mL reduction, therefore other factors could be associated to its efficacy. King et al. (2005) determined the effects of PAA vary and greatly depend on bacteria level and how they are attached to the surface. Nagel et al. (2013) evaluated higher levels of PAA at 400 and 1,000 ppm and reduced *Campylobacter* levels by more than 2.0 log₁₀ cfu/mL.

Scientists have determined a multi-hurdle approach is necessary for adequate pathogen reduction, which uses multiple intervention points throughout processing procedures such as the inside-outside bird washer, brush washer, cabinet washer, or dip tank before and/or after chilling (Smith et al., 2015). Using this approach, the facility does not rely only on a single step intervention, but instead incorporates many applications for reducing foodborne pathogen prevalence prior to entering secondary processing (Stopforth et al., 2007; Zweifel and Stephan, 2012; Sofos et al., 2013). No research was found, however, demonstrating the usefulness of consecutive, sequential

hurdles for pathogen reduction. This study demonstrates the possibility that consecutive application of chemicals in a specific sequential order may reduce *Campylobacter* prevalence.

Conclusions

Findings from this study demonstrated PoultrypHresh™ and PAA could reduce the prevalence of *Campylobacter* on poultry parts when used separately. However, when evaluated sequentially, PAA followed by PoultrypHresh™ produced the best results and significantly reduced pathogen presence compared to other treatments. Such findings may be extremely beneficial and lead to better intervention strategies in future antimicrobial applications.

References

- Allos, B. 2001. *Campylobacter jejuni* infections: update on emerging issues and trends. Clin. Infectious. Dis. 32(8):1201-1206.
- Bacon, R. T., K. E. Belk, J. N. Sofos, R. P. Clayton, J. O. Reagan, and G. C. Smith. 2000. Microbial populations on animal hides and beef carcasses at different stages of slaughter in plants employing multiple-sequential interventions for decontamination. J. Food Prot. 63:1080-1086.
- Bauermeister, L. J. 2015. Evaluation of poultry meat safety and quality using peracetic acid in poultry chillers. Dissertation Auburn University.
- Bauermeister, L. J., J. W. Bowers, J. C. Townsend, and S. R. McKee. 2008. Validating

- the efficacy of peracetic acid mixture as an antimicrobial in poultry chillers. *J. Food Prot.* 71:1119-1122.
- Bennett, P. 2008. Compliance guidelines for controlling *Salmonella* and *Campylobacter* in poultry. 2nd ed. http://www.fsis.usda.gov/wps/wcm/connect/1d562776-ebfb4ea5-a19b-e994776a02c6/Compliance_Guideline_Controller_Salmonella_Poultry.pdf?MOD=AJPERES.
- Black, R. E., M. M. Levine, M. L. Clements, T. P. Hughs, and M. J. Blaser. 1988. Experimental *Campylobacter jejuni* infection in humans. *J. Infect. Dis.* 157:472-479.
- Cox, N. A., L. J. Richardson, M. E. Berrang, P. J. Fedorka-Cray, and R. J. Buhr. 2009. *Campylobacter coli* naturally resistant to elevated levels of gentamicin as a marker strain in poultry research. *J. Food Prot.* 72:1288-1292.
- Food Safety and Inspection Service (FSIS). 2015. Pathogen Reduction - *Salmonella* and *Campylobacter* Performance Standards Verification Testing. United States Department of Agriculture.
- FSIS Directive. 2018. Safe and suitable ingredients used in the production of meat, poultry, and egg products. FSIS Directive 7120.1 Rev. 46. Washington, DC.
- Gill, C. O., and M. Badoni. 2004. Effects of peroxyacetic acid, acidified sodium chlorite or lactic acid solutions on the microflora of chilled beef carcasses. *Int. J. Food Microbiol.* 91:43-50.
- Harrison, W. A., C. J. Griffith, D. Tennant, and A. C. Peters. 2001. Incidence of *Campylobacter* and *Salmonella* isolated from retail chicken and associated packaging in South Wales. *Lett. Appl. Microbiol.* 33:450-454.

- Humphrey, T., S. O'Brien, and M. Madsen. 2007. *Campylobacters as zoonotic pathogens: a food production perspective*. *Int. J. Food Microbiol.* 117:237-257.
- Inspection Methods. 2018. *Pathogen reduction - Salmonella and Campylobacter performance standards verification testing*. Food Safety Inspection Service (FSIS). <https://www.fsis.usda.gov/>.
- King, D. A., L. M. Lucia, A. Castillo, G. R. Acuff, K. B. Harris, and J. W. Savell. 2005. *Evaluation of peracetic acid as a post-chilling intervention for control of Escherichia coli 0157:H7 and Salmonella Typhimurium on beef carcass surfaces*. *Meat Sci.* 69:401-407.
- Landrum, M. A., N. A. Cox, D. E. Cosby, M. E. Berrang, and S. M. Russell. 2016. *Treatment with a low pH processing aid to reduce Campylobacter counts on broiler parts*. *Poult. Sci.* 96(4):1028-1031.
- Moore, J. E., T. S. Wilson, D. R. Wareing, T. J. Humphrey, and P. G. Murphy. 2002. *Prevalence of thermophilic Campylobacter spp. in ready-to-eat foods and raw poultry in Northern Ireland*. *J. Food Prot.* 65:1326-1328.
- Nagel, G. M., L. J. Bauermeister, C. L. Bratcher, M. Singh, and S. R. McKee. 2013. *Salmonella and Campylobacter reduction and quality characteristics of poultry carcasses treated with various antimicrobials in a post-chill immersion tank*. *Int. J. Food Prot.* 165:281-286.
- Oyarzabal, O. A. 2005. *Reduction of Campylobacter spp. by commercial antimicrobials applied during the processing of broiler chickens: a review from the United States perspective*. *J. Food Prot.* 68(8):1752-1760.
- Robinson, D. A. 1981. *Infective dose of Campylobacter jejuni in milk*. *Br. Med. J.*

282:1584.

- Smith, J., S. Corkran, S. R. McKee, S. F. Bilgili, and M. Singh. 2015. Evaluation of post chill applications of antimicrobials against *Campylobacter jejuni* on poultry carcasses. *J. Appl. Poult. Res.* 24:451-456.
- Sofos, J. N., G. Flick, G. Nychas, C. A. O'Bryan, S. C. Ricke, and P. G. Crandall. 2013. Meat, Poultry and Seafood. Pages 111-167 in *Food Microbiology: Fundamentals and Frontiers*. 4th ed. M. P. Doyle, and R. L. Buchanan, eds. ASM Press. Washington, DC.
- Stern, N. J., B. Wojton, and K. Kwiatek. 1992. A differential selective medium and dry ice generated atmosphere for recovery of *Campylobacter jejuni*. *J. Food Prot.* 55:514-517.
- Stopforth, J. D., R. O'Connor, M. Lopes, B. Kottapalli, W. E. Hill, and M. Samadpour. 2007. Validation of individual and multiple-sequential interventions for reduction of microbial populations during processing of poultry carcasses and parts. *J. Food Prot.* 70:1393-1401.
- Suktet, N., P. Tuitemwong, K. Tuitemwong, W. Poonlapdecha, and L. E. Erickson. 2017. Inactivation of *Campylobacter* during immersion chilling of chicken carcasses. *J. Food Eng.* 202:25-33.
- USDA-FSIS. 2016. Sanitation performance standards compliance guide. Title 9. Code of Federal Regulations 416.
- Zweifel, C., and R. Stephan. 2012. Microbial decontamination of poultry carcasses. Pages 60-95 in *Microbial Decontamination in the Food Industry*. A. Demirci, and M. O. Ngadi, eds. Woodhead Publishing, Cambridge, UK.

Table 6.1. Average Log₁₀ cfu/mL of *C. coli* recovered by replicate from sequentially dip treated thighs with no treatment, water-water, PoultrypHresh™-PoultrypHresh™, Peracetic acid-Peracetic acid, PoultrypHresh™-Peracetic acid, or Peracetic acid-PoultrypHresh™ (mean ± standard error) in 5 study replications. ^{a, b, c, d, e} values with different subscripts are significantly ($P \leq 0.05$) different by Tukey's Honest Significant Difference test in skin-on thighs.

Treatment		Average Log ₁₀ cfu/mL	Reduction from Untreated (%)	Reduction from Water (%)
1 st Dip	2 nd Dip			
Untreated	-	5.64 ^a ± 0.04	-	-
Water	Water	4.87 ^b ± 0.05	83.0	-
PpH ¹	PpH	3.90 ^c ± 0.06	98.2	89.3
PAA ²	PAA	3.50 ^d ± 0.08	99.3	95.7
PpH	PAA	3.53 ^d ± 0.08	99.2	95.4
PAA	PpH	2.52 ^e ± 0.09	99.9	99.6

¹ PpH = CMS PoultrypHresh™

² PAA = peracetic acid

CHAPTER 7
CONCLUSIONS

Campylobacter is the source cause of gastroenteritis causing fever, abdominal pain and diarrhea in roughly 850,000 individuals in the United States each year (USDA, 2017). *Campylobacter* is known to be commonly transmitted through poultry and poultry products and can greatly impact the industry (Allos, 2001; Kessel et al., 2001; Cox et al., 2010). One reason *Campylobacter* is so dangerous is the low infectious dose, as little as 500 organisms, required for illness to occur (Robinson, 1981; Black et al., 1988; Friedman et al., 2000). The USDA-FSIS has attempted to minimize the incidence of *Campylobacter* by making performance standards stricter in poultry processing facilities. It has become essential for processors to find the most effective treatments to reduce or eliminate *Campylobacter* in processing facilities. There are many antimicrobials approved for use in poultry processing, although few are used commercially (Dickson and Anderson, 1992; Corry et al., 1995). Many factors determine whether a chemical is ideal as an intervention treatment including documented efficacy, appropriate concentration, contact time, cost efficiency, and whether it damages the product or processing equipment (Bauermeister et al., 2008).

This research focused on evaluating a low pH processing aid, CMS PoultryHresh™, for reducing the prevalence of *Campylobacter* on poultry. The studies evaluated whole carcasses, parts (split breasts and thighs), and livers naturally contaminated and inoculated with a marker strain *C. coli* resistant to gentamicin (Cox et al., 2009). Using whole carcasses, parts and livers demonstrated the many possible intervention points PoultryHresh™ could be beneficial for use. It also shows the effectiveness on different surface types, smooth surfaces (i.e. the liver) and porous surfaces (i.e. poultry skin). Many researchers have determined bacteria's ability to

become lodged and proliferate in the skin's layers (Thomas and McMeekin, 1980; Lillard, 1989; FAO, 2009). Pathogens firmly attached or entrapped in the skin become more difficult for antimicrobials to reduce or remove. These studies showed significant reductions on either type of surface, therefore skin did not appear to be a factor in the effectiveness of PoultrypHresh™. Each study showed dip treatments significantly reduce *Campylobacter* prevalence when compared to untreated or water treated groups.

In whole carcasses naturally contaminated with *Campylobacter*, the incidence was lowered from 83.6% in untreated and 69.4% in water treated carcasses to only 5.6% using a 20 second PoultrypHresh™ dip. Parts were then evaluated using a 25 second dip of PoultrypHresh™, reducing *Campylobacter* on split breast by 99.6% and 99.4% on thighs when compared to untreated parts. Surfactant was added to PoultrypHresh™ in an additional treatment, PoultrypHresh™ Plus, on livers and significantly increased *Campylobacter* reductions. Compared to untreated livers, a 15 second dip in PoultrypHresh™ reduced *C. coli* by 98.1% and PoultrypHresh™ Plus by 99.4%. The final study evaluated consecutive dips on thighs of either PoultrypHresh™ or peracetic acid (PAA), an organic peroxide currently being used in a variety of processing facilities. Other treatments dipped sequentially in PoultrypHresh™ and PAA, demonstrating some very interesting results. Thighs treated in PoultrypHresh™ followed by PAA showed significant *Campylobacter* reductions compared to untreated and water treated samples. However, when treated with PAA followed by PoultrypHresh™ significantly greater reductions were found compared to all other treatment combinations evaluated, reducing *Campylobacter* prevalence in untreated by 99.9% and 99.6% from water treated thighs.

Previous research has shown multiple intervention points are necessary to significantly reduce foodborne pathogens prior to them reaching secondary processing (Stopforth et al., 2007; Zweifel and Stephan, 2012; Sofos et al., 2013). This research demonstrated PoultrypHresh™ can significantly reduce *Campylobacter* when used in only a single intervention point. If it was implemented at multiple intervention points, further reductions or elimination of pathogens may be found. Sequential dips treating with chemicals in sequence may alter the presence of pathogens. This may benefit future research and allow new intervention strategy opportunities. Future studies should further investigate sequential dipping at various concentrations and treatment times to enhance reductions. Therefore, overall this research shows PoultrypHresh™ may be beneficial for processors to meet regulatory requirements and reduce the occurrence of campylobacteriosis worldwide being used at various intervention points throughout poultry processing. It also demonstrates the possibility of coupling it with other common chemicals in sequence could cause significant pathogen reductions.

References

- Allos, B. 2001. *Campylobacter jejuni* infections: update on emerging issues and trends. Clin. Infect. Dis. 32(8):1201-1206.
- Bauermeister, L. J., J. W. Bowers, J. C. Townsend, and S. R. McKee. 2008. The microbial and quality properties of poultry carcasses treated with peracetic acid as an antimicrobial treatment. Poult. Sci. 87:2390-2398.
- Black, R. E., M. M. Levine, M. L. Clements, T. P. Hughs, and M. J. Blaser. 1988.

- Experimental *Campylobacter jejuni* infection in humans. *J. Infect. Dis.* 157:472-479.
- Corry, J., C. James, S. James, and M. Hinton. 1995. *Salmonella*, *Campylobacter* and *Escherichia coli* 0157:H7 decontamination techniques for the future. *Int. J. Food Microbiol.* 28:187-196.
- Cox, N. A., L. J. Richardson, M. E. Berrang, P. J. Fedorka-Cray, and R. J. Buhr. 2009. *Campylobacter coli* naturally resistant to elevated levels of gentamicin as a marker strain in poultry research. *J. Food Prot.* 72(6):1288-1292.
- Cox, N. A., L. J. Richardson, J. A. Cason, R. J. Buhr, Y. Vizzier-Thaxton, D. P. Smith, P. J. Fedorka-Cray, C. P. Romanenghi, L. V. B. Pereira, and M. P. Doyle. 2010. Comparison of neck skin excision and whole carcass rinse sampling methods for microbiological evaluation of broiler carcasses before and after immersion chilling. *J. Food Prot.* 73(5):976-980.
- Dickson, J. S., and M. E. Anderson. 1992. Microbiological decontamination of food animal carcasses by washing and sanitizing systems: a review. *J. Food Prot.* 55:133-140.
- FAO [Food and Agriculture Organization of the United Nations]. 2009. *Salmonella* and *Campylobacter* in chicken meat: meeting report. Page 56 in *Microbiological Risk Assessment Series No. 19*. Rome.
- Friedman, C. R., J. Neimann, H. C. Wegener, and R. V. Tauxe. 2000. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. Pages 121–138 in *American Society for Microbiology Press. I. Nachamkin, and M. J. Blaser, eds.* Washington, D.C.

- Kessel, A. S., I. A. Gillespie, S. J. O'Brien, G. K. Adak, T. J. Humphrey, and L. R. Ward. 2001. General outbreaks of infectious intestinal disease linked with poultry, England and Wales, 1992-1999. *Commun. Dis. Public Health*. 4:171-177.
- Lillard, H. S. 1989. Incidence and recovery of salmonellae and other bacteria from commercially processed poultry carcasses at selected pre- and post-evisceration steps. *J. Food Prot.* 52:88-91.
- Robinson, D. A. 1981. Infective dose of *Campylobacter jejuni* in milk. *Br. Med. J.* 282:1584.
- Sofos, J. N., G. Flick, G. Nychas, C. A. O'Bryan, S. C. Ricke, and P. G. Crandall. 2013. Meat, Poultry and Seafood. Pages 111-167 in *Food Microbiology: Fundamentals and Frontiers*. 4th ed. M. P. Doyle, and R. L. Buchanan, eds. ASM Press. Washington, D.C.
- Stopforth, J. D., R. O'Connor, M. Lopes, B. Kottapalli, W. E. Hill, and M. Samadpour. 2007. Validation of individual and multiple-sequential interventions for reduction of microbial populations during processing of poultry carcasses and parts. *J. Food Prot.* 70(6):1393-1401.
- Thomas, C. J., and T. A. McMeekin. 1980. Contamination of broiler carcass skin during commercial processing procedures: an electron microscopic study. *Appl. Environ. Microbiol.* 40:133-144.
- United States Department of Agriculture (USDA). 2017. Cost estimates of foodborne illnesses. <https://www.ers.usda.gov/data-products/cost-estimates-of-foodborne-illnesses/>.
- Zweifel, C., and R. Stephan. 2012. Microbial decontamination of poultry carcasses. Pages

60-95 in *Microbial Decontamination in the Food Industry*. A. Demirci, and M. O. Ngadi, eds. Woodhead Publishing, Cambridge, UK.

CHAPTER 8
REFERENCES

- Acuff, G. R., C. Vanderzant, M. O. Hanna, J. G. Ehlers, F. A. Golan, and F. A. Gardner. 1986. Prevalence of *Campylobacter jejuni* in turkey carcass processing and further processing of turkey products. *J. Food Prot.* 49(9):712-717.
- Alakomi, H. L., E. Skytta, M. Saarela, T. Mattila-Sandholm, K. Latva-Kala, and I. M. Helander. 2000. Lactic acid permeabilizes gram-negative bacteria by disrupting the outer membrane. *Appl. Environ. Microbiol.* 66(5):2001-2005.
- Allen, V. M., S. A. Bull, J. E. Corry, G. Domingue, F. Jorgensen, J. A. Frost, R. Whyte, A. Gonzalez, N. Elviss, and T. J. Humphrey. 2007. *Campylobacter* spp. contamination of chicken carcasses during processing in relation to flock colonization. *Int. J. Food Microbiol.* 113(1):54-61.
- Allen, V. M., D. B. Tinker, M. H. Hinton, and C. M. Wathes. 2003. Dispersal of microorganisms in commercial defeathering systems. *Br. Poult. Sci.* 44(1):53-59.
- Allos, B. M. 2001. *Campylobacter jejuni* infections: update on emerging issues and trends. *Clin. Infect. Dis.* 32(8):1201-1206.
- Almanza, A. 2015. Changes to the *Salmonella* and *Campylobacter* verification testing program. Food Safety and Inspection Service, USDA. 80:3940-3950.
- Altekruse, S. F., N. J. Stern, P. I. Fields, and D. L. Swerdlow. 1999. *Campylobacter jejuni* – an emerging foodborne pathogens. *Emerg. Infect. Dis.* 5(1):28-35.
- Anderson, M. E., and R. T. Marshall. 1990. Reducing microbial populations on beef tissues: concentration and temperature of an acid mixture. *J. Food Sci.* 55(4):903.
- Annan-Prah, A., and M. Janc. 1988. The mode of spread of *Campylobacter jejuni/coli* to broiler flocks. *J. Vet. Med.* 35:11-18.

- Anthony, N. B. 1998. A review of genetic practices in poultry: efforts to improve meat quality. *J. Muscle Foods*. 9(1):25-33.
- Arnold, J. W. 2007. Bacterial contamination on rubber picker fingers before, during, and after processing. *Poult. Sci.* 86(12):2671-2675.
- Arnold, J. W., and S. Silvers. 2000. Comparison of poultry processing equipment surfaces for susceptibility to bacterial attachment and biofilm formation. *Poult. Sci.* 79(8):1215-1221.
- Arnold, J. W., and I. E. Yates. 2009. Interventions for control of *Salmonella*: clearance of microbial growth from rubber picker fingers. *Poult. Sci.* 88(6):1292-1298.
- Arritt, F. M. 2000. Efficacy of selected chemicals on the attachment and survival of *Campylobacter jejuni* on chicken breast skin. Master's Thesis. Food Science Technology. Virginia Polytechnic Institute and State University.
- Arritt, F. M., J. D. Eifert, M. D. Pierson, and S. S. Sumner. 2002. Efficacy of antimicrobials against *Campylobacter jejuni* on chicken breast skin. *J. Appl. Poult. Res.* 11(4):358-366.
- Atabay, H. I., and J. E. L. Corry. 1997. The prevalence of Campylobacters and Arcobacters in broiler chickens. *J. Appl. Microbiol.* 83(5):619-626.
- Bacon, R. T., K. E. Belk, J. N. Sofos, R. P. Clayton, J. O. Reagan, and G. C. Smith. 2000. Microbial populations on animal hides and beef carcasses at different stages of slaughter in plants employing multiple-sequential interventions for decontamination. *J. Food Prot.* 63(8):1080-1086.
- Bai, Y., K. R. Coleman, C. W. Coleman, and A. L. Waldroup. 2007. Effect of

- cetylpyridinium chloride (Cecure CPC Antimicrobial) on the refrigerated shelf life of fresh boneless, skinless broiler thigh meat. *Int. J. Poult. Sci.* 6(4):91-94.
- Bailey, J. S., J. E. Thomson, and N. A. Cox. 1987. Contamination of poultry during processing. Pages 193-211 in the *Microbiology of Poultry Meat Products*. F. E. Cunningham and N. A. Cox, eds. Academic Press, Orlando, FL.
- Bailey, J. S., J. E. Thomson, N. A. Cox, and A. D. Shackelford. 1986. Chlorine spray washing to reduce bacterial contamination of poultry processing equipment. *Poult. Sci.* 65(6):1120-1123.
- Baker, R. C., and C. A. Bruce. 1995. Further processing of poultry. In *Processing of Poultry*. G. C. Mead, ed. Springer. Boston, MA.
- Baldry, M. G. C., and J. A. L. Fraser. 1988. Disinfection with peroxygens. In *Industrial Biocides: reports on applied chemistry*. K. R., Payne, ed. John Wiley and Sons. Hoboken, N.J.
- Barbut, S. 2002. *Poultry products processing*. CRC Press. Boca Raton, FL.
- Bashor, M. P., P. A. Curtis, K. M. Keener, B. W. Sheldon, S. Kathariou, and J. A. Osborne. 2004. Effects of carcass washers on *Campylobacter* contamination in large broiler processing plants. *Poult. Sci.* 83:1232-1239.
- Batz, M. B., S. Hoffmann, and J. G. Morris. 2012. Ranking the disease burden of 14 pathogens in food sources in the United States using attribution data from outbreak investigations and expert elicitation. *J. Food Prot.* 75(7):1278-1291.
- Bauermeister, L. J. 2015. Evaluation of poultry meat safety and quality using peracetic acid in poultry chillers. Dissertation Auburn University.
- Bauermeister, L. J., J. W. Bowers, J. C. Townsend, and S. R. McKee. 2008a. The

- microbial and quality properties of poultry carcasses treated with peracetic acid as an antimicrobial treatment. *Poult. Sci.* 87(11):2390-2398.
- Bauermeister, L. J., J. W. Bowers, J. C. Townsend, and S. R. McKee. 2008b. Validating the efficacy of peracetic acid mixture as an antimicrobial in poultry chillers. *J. Food Prot.* 71:1119-1122.
- Bautista, D. A., N. Sylvester, S. Barbut, and M. W. Griffiths. 1997. The decontamination efficacy of antimicrobial rinses on turkey carcasses using response surface designs. *Int. J. Food Microbiol.* 34(3):279-292.
- Beers K., J. Rheingans, K. Chinault, P. Cook, B. Smith, and A. Waldroup. 2006. Microbial efficacy of commercial application of Cecure CPC antimicrobial to ingesta-contaminated pre-chill broiler carcasses. *Int. J. Poult. Sci.* 5(8):698-703.
- Behravesh, C. B., I. T. Williams, and R. V. Tauxe. 2012. Emerging foodborne pathogens and problems: expanding prevention efforts before slaughter or harvest. Institute of Medicine. Improving food safety through a one-health approach: workshop summary. National Academics Press (US). Washington, D.C.
- Bell, K. Y., C. N. Cutter, and S. S. Sumner. 1997. Reduction of foodborne microorganisms on beef carcass tissue using acetic acid, sodium bicarbonate, and hydrogen peroxide spray washes. *Food Microbiol.* 14(5):439-448.
- Bender, F. G., and E. Brotsky. 1992. Process for treating poultry carcasses to control *Salmonella* growth. U.S. Patent 5,143,739.
- Bennett, P. 2008. Compliance guidelines for controlling *Salmonella* and *Campylobacter* in poultry. 2nd ed. http://www.fsis.usda.gov/wps/wcm/connect/1d562776-ebfb4ea5-a19b-e994776a02c6/Compliance_Guideline_Controller_Salmonella_

Poultry.pdf.

- Berndtson, E., M. L. Danielsson-Tham, and A. Engvall. 1996. *Campylobacter* incidence on a chicken farm and the spread of *Campylobacter* during the slaughter process. *Int. J. Food Microbiol.* 32:35-47.
- Berndtson, E., and A. Engvall. 1989. Epidemiological studies of factors affecting the occurrence of campylobacters in broiler farms. Pages 61-62 in the Proceedings of the fifth international workshop on *Campylobacter* infections. G. M. Ruiz-Palacios, E. Calva, B. R. Ruiz-Palacios, eds. Puerto Vallarta, Mexico.
- Berrang, M. E., R. J. Buhr, J. A. Cason, and J. A. Dickens. 2001. Broiler carcass contamination with *Campylobacter* from feces during defeathering. *J. Food Prot.* 64(12):2063-2066.
- Berrang, M. E., N. A. Cox, G. R. Gamble, R. J. Meinersmann, B. B. Oakley, and A. Hinton, Jr. 2018. Contributors to the 100-fold increase in *Campylobacter* numbers on broiler carcasses during defeathering. *Poultry USA*. Watt Publishing Company. In Press.
- Berrang, M. E., and J. A. Dickens. 2000. Presence and level of *Campylobacter* spp. on broiler carcasses throughout the processing plant. *J. Appl. Poult. Res.* 9(1):43-47.
- Berrang, M. E., R. J. Meinersmann, D. P. Smith, and H. Zhuang. 2008. The effect of chilling in cold air or ice water on the microbiological quality of broiler carcasses and the population of *Campylobacter*. *Poult. Sci.* 87(5):992-998.
- Berrang, M. E., W. R. Windham, and R. J. Meinersmann. 2011. *Campylobacter*, *Salmonella*, and *Escherichia coli* on broiler carcasses subjected to a high pH scald and low pH postpick chlorine dip. *Poult. Sci.* 90(4):896-900.

- Bilgili, S. F., D. E. Conner, J. L. Pinion, and K. C. Yamblyn. 1998. Broiler skin color as affected by organic acids: influence of concentration and method of application. *Poult. Sci.* 77(5):752-757.
- Bilgili, S. F., A. L. Waldroup, D. Zelenka, and J. E. Marion. 2002. Visible ingesta on pre-chill carcasses does not affect the microbiological quality of broiler carcasses after immersion chilling. *J. Appl. Poult. Res.* 11(3):233-238.
- Black, R. E., M. M. Levine, M. L. Clements, T. P. Hughs, and M. J. Blaser. 1988. Experimental *Campylobacter jejuni* infection in humans. *J. Infect. Dis.* 157(3):472-479.
- Blaser, M. J. 1997. Epidemiologic and clinical features of *Campylobacter jejuni* infections. *J. Infect. Dis.* 176(Suppl. 2):S103-S105.
- Bolton, F. J., D. Coates, D. N. Hutchinson, and A. F. Godfree. 1987. A study of thermophilic campylobacters in a river system. *J. Appl. Bacteriol.* 62(2):167-176.
- Bolton, F. J., P. M. Hinchliffe, D. Coates, and L. Robertson. 1982. A most probable number method for estimating small numbers of campylobacters in water. *J. Hyg.* 89(2):185-190.
- Brennhovd, O., G. Kapperud, and G. Langeland. 1992. Survey of thermotolerant *Campylobacter* spp. and *Yersinia* spp. in three surface water sources in Norway. *Int. J. Food Microbiol.* 15:327-338.
- Brinez, W. J., A. X. Roig-Sagues, M. M. Hernandez Herrero, T. Lopez-Pedemonte, and B. Guamis. 2006. Bactericidal efficacy of peracetic acid in combination with hydrogen peroxide against pathogenic and non-pathogenic strains of

- Staphylococcus* spp., *Listeria* spp., and *Escherichia coli*. Food Contr. 17(7):516-521.
- Brown, H. G. 2003. Microbial problems, causes, and solutions in meat and poultry processing operations. Pages 89-93 in the Proceedings of the 56th American Meat Science Association Reciprocal Meat Conference. Columbia, MI.
- Bryan, F. L., and M. P. Doyle. 1995. Health risks and consequences of *Salmonella* and *Campylobacter jejuni* in raw poultry. J. Food Prot. 58:326-344.
- Buhr, R. J., N. A. Cox, N. J. Stern, M. T. Musgrove, J. L. Wilson, and K. L. Hiatt. 2002. Recovery of *Campylobacter* from segments of the reproductive tract of broiler breeder hens. Avian Dis. 46(4):919-924.
- Buswell, C. M., Y. M. Herlihy, L. M. Lawrence, J. T. M. McGuiggan, P. D. Marsh, C. W. Keevil, and S. A. Leach. 1998. Extended survival and persistence of *Campylobacter* spp. in water and aquatic biofilms and their detection by immunofluorescent-antibody and rRNA staining. Appl. Environ. Microbiol. 64(2):733-741.
- Butzler, J. P. 2004. *Campylobacter*, from obscurity to celebrity. Clin. Microbiol. Infect. 10(10):868-876.
- Capita, R., C. Alonso-Calleja, M. C. Garcia-Fernandez, and B. Moreno. 2002. Review: trisodium phosphate (TSP) treatment for decontamination of poultry. Food Sci. Technol. Int. 8(1):11-24.
- Carciofi, B. A. M., and J. B. Laurindo. 2007. Water uptake by poultry carcasses during cooling by water immersion. Chem. Eng. Process. 46(5):444-450.
- Carmarda, A., D. G. Newell, R. Nasti, and G. Di Modugno. 2000. Genotyping

- Campylobacter jejuni* strains isolated from the gut and oviduct of laying hens. Avian Dis. 44:907-912.
- Carroll, C. D., and C. Z. Alvarado. 2008. Comparison of air and immersion chilling on meat quality and shelf life of marinated broiler breast fillets. Poult. Sci. 87:368-372.
- Cason, J. A., N. A. Cox, R. J. Buhr, and L. J. Richardson. 2010. Comparison of the statistics of *Salmonella* testing of chilled broiler chicken carcasses by whole-carcass rinse and neck skin excision. Poult. Sci. 89(9):2038-2040.
- Cason, J. A., and A. Hinton, Jr. 2006. Coliforms, *Escherichia coli*, *Campylobacter* and *Salmonella* in a counterflow poultry scalding tank with a dip tank. Int. J. Poult. Sci. 5(9):846-849.
- Cason, J. A., A. Hinton, Jr., and K. D. Ingram. 2000. Coliform, *Escherichia coli*, and salmonellae concentrations in a multiple-tank, counterflow poultry scalding tank. J. Food Prot. 63(9):1184-1188.
- Centers for Disease Control and Prevention. 1993. Update: multistate outbreak of *Escherichia coli* O157:H7 infections from hamburgers, western United States, 1992-1993. Morb. Mortal. Wkly Rep. 42(14):258-263.
- Centers for Disease Control and Prevention. 2000. National Antimicrobial Resistance Monitoring System (NARMS) – 1999 annual report. <http://www.cdc.gov/narms>.
- Centers for Disease Control and Prevention. 2002. Preliminary FoodNet data on the incidence of infection with pathogens transmitted commonly through food - selected sites, United States, 2001. Morb. Mortal. Wkly Rep. 51(15):325-329.
- Centers for Disease Control and Prevention (CDC). 2013. Multistate outbreak of

- Campylobacter jejuni* infections associated with undercooked chicken livers-northeastern United States 2012. *Morb. Mortal. Wkly Rep.* 62:874-876.
- Chantarapanont, W., M. E. Berrang, and J. F. Frank. 2004. Direct microscopic observation of viability of *Campylobacter jejuni* on chicken skin treated with selected chemical sanitizing agents. *J. Food Prot.* 67(6):1146-1152.
- Chuma, T., T. Yamada, K. Yano, K. Okamoto, and H. Yugi. 1994. A survey of *Campylobacter jejuni* in broilers from assignment to slaughter using DNA-DNA hybridization. *J. Vet. Med. Sci.* 56(4):697-700.
- CIA World Factbook. 2017. <https://www.cia.gov/library/publications/the-world-factbook/>.
- Clarke, A. G., and D. H. Bueschkens. 1985. Laboratory infection of chicken eggs with *Campylobacter jejuni* by using temperature or pressure differentials. *Appl. Environ. Microbiol.* 49(6):1467-1471.
- Cloak, O. M., G. Duffy, J. J. Sheridan, I. S. Blair, and D. A. McDowell. 2001. A survey on the incidence of *Campylobacter* spp. and the development of a surface adhesion polymerase chain reaction (SA-PCR) assay for the detection of *Campylobacter jejuni* in retail meat products. *Food Microbiol.* 18(3):287-298.
- Conner, D. E., and S. F. Biligli. 1994. Skin attachment model for improved laboratory evaluation of potential carcass disinfectants for their efficacy against *Salmonella* attached to broiler skin. *J. Food Prot.* 57(8):684-688.
- Coppen, P. P. 1993. *Salmonella* reduction in poultry processing - application of TSP process to European industry. Pages 524-531 in Proceedings of the 11th European Symposium on the Quality of Poultry Meat. Branche Francaise.

- Corry, J. E., C. James, S. J. James, and M. Hinton. 1995. *Salmonella*, *Campylobacter* and *Escherichia coli* 0157:H7 decontamination techniques for the future. *Int. J. Food Microbiol.* 28(2):187-196.
- Cox, N. A., J. S. Bailey, L. J. Richardson, R. J. Buhr, D. E. Cosby, J. L. Wilson, K. L. Hiatt, G. R. Siragusa, and D. V. Bourassa. 2005b. Presence of naturally occurring *Campylobacter* and *Salmonella* in the mature and immature ovarian follicles of late-life broiler breeder hens. *Avian Dis.* 49(2):285-287.
- Cox, N. A., C. L. Hofacre, J. S. Bailey, R. J. Buhr, J. L. Wilson, K. L. Hiatt, L. J. Richardson, M. T. Musgrove, D. E. Cosby, J. D. Tankson, Y. L. Vizzier, P. F. Cray, L. E. Vaughn, P. S. Holt, and D. V. Bourassaa. 2005a. Presence of *Campylobacter jejuni* in various organs one hour, one day, and one week following oral or intracloacal inoculation of broiler chicks. *Avian Dis.* 49(1):155-158.
- Cox, N. A., L. J. Richardson, M. E. Berrang, P. J. Fedorka-Cray, and R. J. Buhr. 2009a. *Campylobacter coli* naturally resistant to elevated levels of gentamicin as a marker strain in poultry research. *J. Food Prot.* 72(6):1288-1292.
- Cox, N. A., L. J. Richardson, R. J. Buhr, and P. J. Fedorka-Cray. 2009b. *Campylobacter* species occurrence within internal organs and tissues of commercial caged leghorn laying hens. *Poult. Sci.* 88(11):2449-2456.
- Cox, N. A., L. J. Richardson, R. J. Buhr, P. J. Fedorka-Cray, J. S. Bailey, J. L. Wilson, and K. L. Hiatt. 2006. Natural presence of *Campylobacter* spp. in various internal organs or commercial broiler breeder hens. *Avian Dis.* 50(3):450-453.
- Cox, N. A., L. J. Richardson, R. J. Buhr, J. K. Northcutt, J. S. Bailey, P. J. Fedorka

- Cray, and K. L. Hiatt. 2007. Recovery of *Campylobacter* and *Salmonella* serovars from the spleen, liver/gallbladder, and ceca of six and eight week old commercial broilers. *J. Appl. Poult. Res.* 16(4):477-480.
- Cox, N. A., L. J. Richardson, J. A. Cason, R. J. Buhr, Y. Vizzier-Thaxton, D. P. Smith, P. J. Fedorka-Cray, C. P. Romanenghi, L. V. Pereira, and M. P. Doyle. 2010. Comparison of neck skin excision and whole carcass rinse sampling methods for microbiological evaluation of broiler carcasses before and after immersion chilling. *J. Food Prot.* 73(5):976-980.
- Cox, N. A., N. J. Stern, J. L. Wilson, M. T. Musgrove, R. J. Buhr, and K. L. Hiatt. 2002. Isolation of *Campylobacter* spp. from semen samples of commercial broiler breeder roosters. *Avian Dis.* 46(3):717-720.
- Curtis, P., and J. Butler. 2009. Controlling *Campylobacter* in poultry plants. Auburn University. <http://www.fsis.usda.gov/>.
- Davies, A., and R. Board. 1998. The microbiology of meat and poultry. 1st ed. A. Davis and R. Board, eds. Blackie Academic and Professional. London, UK.
- Demirok, E., G. Veluz, W. V. Stuyvenberg, M. P. Castaneda, A. Byrd, and C. Z. Alvarado. 2013. Quality and safety of broiler meat in various chilling systems. *Poult. Sci.* 92(4):1117-1126.
- Deumier, F. 2004. Decontamination of deboned chicken legs by vacuum tumbling in lactic acid solution. *Int. J. Food Sci. Tech.* 41(1):23-32.
- DeWaal, C. S., C. Roberts, and C. Catella. 2012. Antibiotic resistance in foodborne

pathogens: evidence of the need for risk management strategy. Center for Science in the Public Interest, White Paper. http://cspinet.org/new/pdf/abrfoodborne_nepathogenswhitepaper.pdf.

- Dickens, J. A., N. A. Cox, J. S. Bailey, and J. E. Thomson. 1985. Automated microbiological sampling of broiler carcasses. *Poult. Sci.* 64:1116-1120.
- Dickens, J. A., B. G. Lyon, A. D. Whittemore, and C. E. Lyon. 1994. The effect of an acetic acid dip on carcass appearance, microbiological quality, and cooked breast meat texture and flavor. *Poult. Sci.* 73(4):576-581.
- Dickens, J. A., and A. D. Whittemore. 1995. The effects of extended chilling times with acetic acid on the temperature and microbiological quality of processed poultry carcasses. *Poult. Sci.* 74(6):1044-1048.
- Dickson, J. S., and M. E. Anderson. 1992. Microbiological decontamination of food animal carcasses by washing and sanitizing systems: a review. *J. Food Prot.* 55(2):133-140.
- Dincer, A. H., and A. Unluturk. 2001. Effects of lactic acid and fumaric acids on microbiological stability of turkey breast meat. IX European Symposium on the quality of eggs and egg products and XV European symposium on the quality of poultry meat. Symposium Book.
- Djenane, D., A. Sanchez-Escalante, J. A. Beltran, and P. Roncales. 2003. The shelf life of beef steaks treated with DL-lactic acid and antioxidants and stored under modified atmospheres. *Food Microbiol.* 20(1):1-7.
- Drewniak, E. E., M. A. Howe, Jr., J. E. Goresline, and E. R. Baush. 1954. Studies on

sanitizing methods for use in poultry processing. Circular no. 930. U.S. Department of Agriculture. Washington, D.C.

Dreyfuss, M. S., G. M. Ransom, M. D. Russell, K. E. Barlow, K. M. Pritchard, D. R.

Eblen, C. A. Nadon, P. K. Saini, N. D. O. Antoine, B. E. Rose, and G. W.

Zirnstien. 2007. Pathogen control in meat and poultry production: implementing the USDA food safety and inspection service's hazard analysis and critical control point system. *Infectious Disease: Foodborne Dis.* 15:383-404.

Dubois-Brissonnet, F. 2012. Adaptation of *Salmonella* to antimicrobials in food processing environments. Pages 123-146 in *Salmonella: Distribution, Adaptation, Control Measures and Molecular Technologies*. B. A. Annous, and J. B. Gurtler, eds. Intech.

Edwards, E. S., L. M. Milne, K. Morrow, P. Sheridan, N. Q. Verlander, R. Mulla, J. F.

Richardson, A. Pender, M. Lilley, and M. Reacher. 2014. Campylobacteriosis outbreak associated with consumption of undercooked chicken liver pate in the East of England, September 2011: identification of a dose-response risk. *Epidemiol. Infect.* 142:352-357.

Ellerbroek, L., E. M. Okolocha, and E. Weise. 1996. Lactic acid and trisodium phosphate for decontamination of poultry meat. Pages 187-195 in *Microbial Methods for the Meat Industry*. M. H. Hinton, and C. Rowlings, eds. University of Bristol Press. Bristol.

Engberg, J., F. M. Aarestrup, D. E. Taylor, P. Gerner-Smidt, and I. Nachamkin. 2001.

- Quinolone and macrolide resistance in *Campylobacter jejuni* and *C. coli*: resistance mechanisms and trends in human isolates. *Emerg. Infect. Dis.* 7(1):24-34.
- Evans, S. J., and A. R. Sayers. 2000. A longitudinal study of *Campylobacter* infection of broiler flocks in Great Britain. *Prev. Vet. Med.* 46(3):209-223.
- FAO. 2003. Risk assessment of *Campylobacter* spp. in broiler chickens and *Vibrio* spp. in seafood. Report of a Joint FAO/WHO Expert Consultation. Bangkok, Thailand.
- FAO [Food and Agriculture Organization of the United Nations]. 2009. *Salmonella* and *Campylobacter* in chicken meat: meeting report. Page 56 in Microbiological Risk Assessment Series No. 19. Rome.
- FDA. Federal Register. 2000. Secondary direct additives permitted in food for human consumption. Peroxyacids – Final Rule, 21 CFR Part 173. 65(228):70660-70661.
- FDA. 2015. Food Protection Plan of 2007. <https://www.fda.gov/Food/GuidanceRegulation/FoodProtectionPlan207.default.htm>.
- Federal Register, United States Department of Agriculture. 1982. Meat and poultry products: phosphates and sodium hydroxide. *Fed. Regis.* 47:10779.
- Federal Register, United States Department of Agriculture. 1996. 9 CFR Part 304 et al., Pathogen Reduction; Hazard Analysis and Critical Control Point (HACCP) Systems. Final Rule. 61(144):38846-38848.
- Fernández, H., and V. Pisón. 1996. Isolation of thermotolerant species of *Campylobacter* from commercial chicken livers. *Int. J. Food Microbiol.* 29:75-80.
- Ferrara, O. 2005. Brand preferences among meats: an application of probit models. University of Florida. Master of Science Thesis.

- Firleyanti, A. S., P. L. Connerton, and I. F. Connerton. 2016. Campylobacters and their bacteriophages from chicken liver: The prospect for phage biocontrol. *Int. J. Food Microbiol.* 237:121-127.
- Fletcher, D. L. 2002. Poultry meat quality. *World's Poult Sci. J.* 58(2):131-145.
- Fluckey, W. M., M. X. Sanchez, S. R. Mckee, D. Smith, E. Pendleton, and M. M. Brashears. 2003. Establishment of a microbiological profile for an air-chilling poultry operation in the United States. *J. Food Prot.* 66(2):272-279.
- Food Safety Education - *Campylobacter* question and answers. 2013. United States Department of Agriculture (USDA). <http://www.fsis.usda.gov/>.
- Fortin, N. D. 2003. The hang-up with HACCP: the resistance to translating science into food safety law. *Food Drug Law J.* 58(4):565-593.
- Food Safety and Inspection Service (FSIS). 2015. Pathogen Reduction - *Salmonella* and *Campylobacter* performance standards verification testing. United States Department of Agriculture.
- Food Safety and Inspection Service (FSIS). 2016. New performance standards for *Salmonella* and *Campylobacter* in not-ready-to-eat comminuted chicken and turkey products and raw chicken parts and changes to related agency verification procedures. United States Department of Agriculture. *Fed. Reg.* 81:7285-7300.
- Friedman, C. R., J. Neimann, H. C. Wegener, and R. V. Tauxe. 2000. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. Pages 121–138 in American Society for Microbiology Press. I. Nachamkin, and M. J. Blaser, eds. Washington, D.C.

- FSIS Directive. 2018. Safe and suitable ingredients used in the production of meat, poultry, and egg products. FSIS Directive 7120.1 Rev. 46. Washington, DC.
- Gallo, L., R. E. Schmitt, and W. Schmidt-Lorenz. 1988. Microbial spoilage of refrigerated fresh broilers: I. Bacterial flora and growth during storage. *Food Sci. Tech.* 21(4):216-223.
- Gaudreau, C., and H. Gilbert. 1998. Antimicrobial resistance of clinical strains of *Campylobacter jejuni* subsp. *jejuni* isolated from 1985 to 1997 in Quebec, Canada. *Antimicrob. Agents Chemother.* 42(8):2106-2108.
- Gavin, A., and L. M. Weddig. 1995. Canned foods: principles of thermal process control, acidification, and container closure evaluation. Pages 36-41 in the Food Processors Institute.
- Ge, B., D. G. White, P. F. McDermott, W. Girand, S. Zhao, S. Hubert, and J. Meng. 2003. Antimicrobial-resistant *Campylobacter* species from retail raw meats. *Appl. Environ. Microbiol.* 69(5):3005-3007.
- Geornaras, I., and A. von Holy. 2000. Bacterial counts associated with poultry processing at different sampling times. *J. Basic Microbiol.* 40:343-349.
- Geornaras, I., A. de Jesus, E. van Zyl, and A. von Holy. 1995. Microbiological survey of a South African poultry processing plant. *J. Basic Microbiol.* 35(2):73-82.
- Gibbens, J. C., S. J. Pascoe, S. J. Evans, R. H. Davies, and A. R. Sayers. 2001. A trial of biosecurity as a means to control *Campylobacter* infection of broiler chickens. *Prev. Vet. Med.* 48(2):85-99.
- Giese, J. 1992. Experimental process reduces *Salmonella* on poultry. *Food Technol.* 46:112.

- Giese, J. 1993. *Salmonella* reduction process receives approval. Food Technol. 47:110.
- Gilbert, C. D., Y. Bai, and H. Jiang. 2015. Microbial evaluation of Cecure treated (post chill) raw poultry carcasses and cut-up parts in four commercial broiler processing facilities. Int. J. Poult. Sci. 14(3):120-126.
- Gill, C. O., and M. Badoni. 2004. Effects of peroxyacetic acid, acidified sodium chlorite or lactic acid solutions on the microflora of chilled beef carcasses. Int. J. Food Microbiol. 91(1):43-50.
- Gonzalez-Fandos, E., B. Herrera, and N. Maya. 2009. Efficacy of citric acid against *Listeria monocytogenes* attached to poultry skin during refrigerated storage. Int. J. Food Sci. Technol. 44(2):262-268.
- Gregory, E., H. Barnhart, D. W. Dreesen, N. J. Stern, and J. L. Corn. 1997. Epidemiological study of *Campylobacter* spp. in broilers: source, time of colonization, and prevalence. Avian Dis. 41(4):890-898.
- Guerrero-Legarreta, I. 2010. Handbook of poultry science and technology: secondary processing. John Wiley and Sons, Inc. 2:599-600.
- Gupta, A., J. M. Nelson, T. J. Barrett, R. V. Taxue, S. P. Rossiter, C. R. Friedman, K. W. Joyce, K. E. Smith, T. F. Jones, M. A. Hawkins, B. Shiferaw, J. L. Beebe, D. J. Vugia, T. Rabatsky-Ehr, J. A. Benson, T. P. Root, and F. J. Angulo. 2004. Antimicrobial resistance among *Campylobacter* strains, United States, 1997-2001. Emerg. Infect. Dis. 10(6):1102-1109.
- Hald, B., A. Wedderkopp, and M. Madsen. 2000. Thermophilic *Campylobacter* spp. in Danish broiler production: a cross-sectional survey and a retrospective analysis of risk factors for occurrence in broiler flocks. Avian Pathol. 29(2):123-131.

- Harris, N. V., D. Thompson, D. C. Martin, and C. M. Nolan. 1986. A survey of *Campylobacter* and other bacterial contaminants of pre-market chicken and retail poultry and meats, King County, Washington. *Am. J. Public Health*. 76(4):401-406.
- Harrison, D., J. E. L. Corry, M. A. Tchorzewska, V. K. Morris, and M. L. Hutchison. 2013. Freezing as an intervention to reduce the numbers of campylobacters isolated from chicken livers. *J. Appl. Microbiol.* 57:206-213.
- Harrison, W. A., C. J. Griffith, D. Tennant, and A. C. Peters. 2001. Incidence of *Campylobacter* and *Salmonella* isolated from retail chicken and associated packaging in South Wales. *Lett. Appl. Microbiol.* 33(6):450-454.
- Hiett, K. L., N. J. Stern, P. Fedorka-Cray, N. A. Cox, M. T. Musgrove, and S. Ladely. 2002. Molecular subtype analyses of *Campylobacter* spp. from Arkansas and California poultry operations. *Appl. Environ. Microbiol.* 68(12):6220-6236.
- Havenstein, G. B. 2006. Performance changes in poultry and livestock following 50 years of genetic selection. *Lohmann Info.* 41:30-37.
- Hinton Jr., A., J. A. Cason, M. E. Hume, and K. D. Ingram. 2004a. Use of MIDI-fatty acid methyl ester analysis to monitor the transmission of *Campylobacter* during commercial poultry processing. *J. Food Prot.* 67(8):1610-1616.
- Hinton Jr., A., J. A. Cason, M. E. Hume, and K. D. Ingram. 2004b. Spread of *Campylobacter* spp. during poultry processing in different seasons. *Int. J. Poult. Sci.* 3(7):432-437.
- Hinton Jr., A., J. A. Cason, and K. D. Ingram. 2002. Enumeration and identification of

- yeasts associated with commercial poultry processing and spoilage of refrigerated broiler carcasses. *J. Food Prot.* 65(6):993-998.
- Hinton Jr., A., J. A. Cason, and K. D. Ingram. 2004c. Tracking spoilage bacteria in commercial poultry processing and refrigerated storage of poultry carcasses. *Int. J. Food Microbiol.* 91(2):155-165.
- Hudson, J. A., C. Nicol, J. Wright, R. Whyte, and S. K. Hasell. 1999. Seasonal variation of *Campylobacter* types from human cases, veterinary cases, raw chicken, milk and water. *J. Appl. Microbiol.* 87(1):115-124.
- Hue, O., V. Allain, M. J. Laisney, S. Le Bouquin, F. Lalande, I. Petetin, S. Rouxel, S. Quesne, P. Y. Gloaguen, M. Picherot, J. Santolini, S. Bougeard, G. Salvat, and M. Chemaly. 2011. *Campylobacter* contamination of broiler caeca and carcasses at the slaughterhouse and correlation with *Salmonella* contamination. *Food Microbiol.* 28(5):862-868.
- Huezo, R., D. P. Smith, J. K. Northcutt, and D. L. Fletcher. 2007. Effect of immersion or dry air chilling on broiler carcass moisture retention and breast fillet functionality. *J. Appl. Poult. Res.* 16(3):438-447.
- Huffman, R. D. 2002. Current and future technologies for the decontamination of carcasses and fresh meat. *Meat Sci.* 62:285-294.
- Hui, Y. H., and F. Sherkat. 2005. Biosynthesis of natural aroma compounds. Page 193 in *Handbook of Food Science, Technology, and Engineering*. CRC Press. London, N.Y.
- Humphrey, T., S. O'Brien, and M. Madsen. 2007. *Campylobacters* as zoonotic pathogens: a food production perspective. *Int. J. Food Micro.* 117(3):237-257.

- Hutchinson, M., D. Harrison, I. Richardson, and M. Tchórzewska. 2015. A method for the preparation of chicken liver pâté that reliably destroys *Campylobacters*. *Int. J. Environ. Res. Public Health*. 12:4652-4669.
- Hutchinson, M. L., L. D. Walters, G. C. Mead, M. Howell, and V. M. Allen. 2006. An assessment of sampling methods and microbiological hygiene indicators for process verification in poultry slaughterhouses. *J. Food Prot.* 69(1):145-153.
- Immerseel, F. V., L. De Zutter, K. Houf, F. Pasmans, F. Haesebrouck, and R. Ducatelle. 2009. Strategies to control *Salmonella* in broiler production chain. *World Poult. Sci. J.* 65(3):367-392.
- Inspection Methods. 2018. Pathogen reduction - *Salmonella* and *Campylobacter* performance standards verification testing. Food Safety Inspection Service (FSIS). <https://www.fsis.usda.gov/>.
- International Commission and Microbiological Specifications for Foods (ICMSF). 1998. Microbiological ecology of food commodities. Pages 107-157 in *Poultry and Poultry Products*. Blackie Academic and Professional. London, UK.
- Izat, A. L., M. Colberg, R. A. Thomas, M. H. Adams, and C. D. Driggers. 1990. Effects of lactic acid in processing waters on the incidence of salmonellae on broilers. *J. Food Qual.* 13(4):295-306.
- Izat, A. L., F. A. Gardner, J. H. Denton, and F. A. Golan. 1988. Incidence and level of *Campylobacter jejuni* in broiler processing. *Poult. Sci.* 67(11):1568-1572.
- Jacobs-Reitsma, W. F. 1997. Aspects of epidemiology of *Campylobacter* in poultry. *Vet. Q.* 19(3):113-117.
- Jacobs-Reitsma, W. F., N. M. Bolder, and R. W. A. W. Mulder. 1994. Cecal carriage of

- Campylobacter* and *Salmonella* in Dutch broiler flocks at slaughter: A one-year study. *Poult. Sci.* 73(8):1260-1266.
- Jacobs-Reitsma, W. F., A. W. van de Giessen, N. M. Bolder, and R. W. Mulder. 1995. Epidemiology of *Campylobacter* spp. at two Dutch broiler farms. *Epidemiol. Infect.* 114(3):413-421.
- James, C., C. Vincent, T. I. de Andrade Lima, and S. J. James. 2006. The primary chilling of poultry carcasses - a review. *Int. J. Refrig.* 29(6):847-862.
- Jones, D. M., E. M. Sutcliffe, and A. Curry. 1991. Recovery of viable but non-culturable *Campylobacter jejuni*. *J. Gen. Microbiol.* 137(10):2477-2482.
- Kapperud, G., E. Skjerve, L. Vik, K. Hauge, A. Lysaker, I. Aalmen, S. M. Ostroff, and M. Potter. 1993. Epidemiological investigation of risk factors for *Campylobacter* colonization in Norwegian broiler flocks. *Epidemiol. Infect.* 111(2):245-255.
- Keenan, S., S. Spice, J. Cole, and P. Banfi. 2015. Food safety policy and regulation in the United States. In: European Parliament's Committee on Environment, Public Health and Food Safety (ENVI). Brussels, Belgium. [https://www.europarl.europa.eu/RegData/etudes/STUD/2015/536324/IPOL_STU\(2015\)536324_EN.pdf](https://www.europarl.europa.eu/RegData/etudes/STUD/2015/536324/IPOL_STU(2015)536324_EN.pdf).
- Keener, K. M., M. P. Bashor, P. A. Curtis, B. W. Sheldon, and S. Kathariou. 2004. Comprehensive review of *Campylobacter* and poultry processing. *Compr. Rev. Food Sci. Food Safety.* 3(2):105-116.
- Kemp, G. K., M. L. Aldrich, M. L. Guerra, and K. R. Schneider. 2001. Continuous online processing of fecal- and ingesta- contaminated poultry carcasses using an acidified sodium chlorite antimicrobial intervention. *J. Food Prot.* 64(6):807-812.
- Kemp, G. K., M. L. Aldrich, and A. L. Waldroup. 2000. Acidified sodium chlorite

- antimicrobial treatment of broiler carcasses. *J. Food Prot.* 63(8):1087-1092.
- Kessel, A. S., I. A. Gillespie, S. J. O'Brien, G. K. Adak, T. J. Humphrey, and L. R. Ward. 2001. General outbreaks of infectious intestinal disease linked with poultry, England and Wales, 1992-1999. *Commun. Dis. Public Health.* 4(3):171-177.
- Kim, J. W., S. J. Knabel, and S. Doores. 1993. Penetration of *Salmonella* Typhimurium into turkey skin. *J. Food Prot.* 56(4):292-296.
- Kim, J. W., M. F. Slavik, F. G. Bender. 1994. Removal of *Salmonella* Typhimurium attached to chicken skin by rinsing with trisodium phosphate solution: scanning electron microscopic examination. *J. Food Safety.* 14(1):77-84.
- King, D. A., L. M. Lucia, A. Castillo, G. R. Acuff, K. B. Harris, and J. W. Savell. 2005. Evaluation of peracetic acid as a post-chilling intervention for control of *Escherichia coli* 0157:H7 and *Salmonella* Typhimurium on beef carcass surfaces. *Meat Sci.* 69:401-407.
- Korber, D. R., A. Choi, G. M. Wolfaardt, and D. E. Caldwell. 1996. Bacterial plasmolysis as a physical indicator of viability. *Appl. Environ. Microbiol.* 62(11):3939-3947.
- Landrum, M. A., N. A. Cox, D. E. Cosby, M. E. Berrang, and S. M. Russell. 2016. Treatment with a low pH processing aid to reduce *Campylobacter* on broiler parts. *Poult. Sci.* 96(4):1028-1031.
- Lastovica, A. J. 2006. Emerging *Campylobacter* spp.: the top of the iceberg. *Clin. Microbiol. Newsletter.* 28(7):49-56.
- Lecompte, J. Y., A. Collignan, S. Sarter, E. Cardinale, and A. Kondjoyan. 2009.

- Decontamination of chicken surfaces inoculated with *Listeria innocua*, *Salmonella* Enteritidis, and *Campylobacter jejuni* by contact with a concentrated lactic acid solution. *Br. Poult. Sci.* 50(3):307-317.
- Lee, M. D., S. Sanchez, M. Zimmer, U. Idris, M. E. Berrang, and P. F. McDermott. 2002. Class 1 Integron-associated tobramycin-gentamicin resistance in *Campylobacter jejuni* isolated from the broiler chicken house environment. *Antimicrob. Agents Chemother.* 46(11):3660-3664.
- Leistner, L., and L. G. M. Gorris. 1995. Food preservation by hurdle technology. *Trends Food Sci. Technol.* 6(2):41-46.
- Li, Y., M. F. Slavik, J. T. Walker, and H. Xiong. 1997. Pre-chill spray of chicken carcasses to reduce *Salmonella* Typhimurium. *J. Food Sci.* 62(3):605-607.
- Liem, D. 2002. Personal communication. Environmental Protection Agency: Office of Pesticide Programs, Antimicrobial Division. Washington, D.C.
- Lillard, H. S. 1979. Levels of chlorine and chlorine dioxide of equivalent bactericidal effect in poultry processing water. *J. Food Sci.* 44(6):1594-1597.
- Lillard, H. S. 1980. Effect on broiler carcasses and water of treating chiller water with chlorine or chlorine dioxide. *Poult. Sci.* 59(8):1761-1766.
- Lillard, H. S. 1985. Bacterial cell characteristics and conditions influencing their adhesion to poultry skin. *J. Food Prot.* 48(9):803-807.
- Lillard, H. S. 1986. Distribution of "attached" *Salmonella* Typhimurium cells between poultry skin and a surface film following water immersion. *J. Food Prot.* 49(6):449-454.
- Lillard, H. S. 1988. Effect of surfactant or changes in ionic strength on the attachment of

- Salmonella* Typhimurium to poultry skin and muscle. J. Food Sci. 53(3):727-730.
- Lillard, H. S. 1989. Incidence and recovery of salmonellae and other bacteria from commercially processed poultry carcasses at selected pre- and post-evisceration steps. J. Food Prot. 52(2):88-91.
- Little, C. L., F. J. Gormley, N. Rawal, and J. F. Richardson. 2010. A recipe for disaster: outbreaks of campylobacteriosis associated with poultry liver pâté in England and Wales. Epidemiol. Infect. 138:1691-1694.
- Loretz, M., R. Stephan, and C. Zweifel. 2010. Antimicrobial activity of decontamination treatments for poultry carcasses: a literature survey. Food Control. 21(6):791-804.
- Man, S. M. 2011. The clinical importance of emerging *Campylobacter* species. Nat. Rev. Gastroenterol. Hepatol. 8(12):669-685.
- Marriott, N. 2013. Principles of Food Sanitation. 3rd ed. Springer Science & Business Media.
- Mbata, T. 2017. Poultry meat pathogens and its control. Int. J. Food Safety. 7:20-28.
- McCarthy, N., and J. Giesecke. 2001. Incidence of Guillain-Barre Syndrome following infection with *Campylobacter jejuni*. Am. J. Epidemiol. 153(6):610-614.
- McKee, S. 2001. Chilling of poultry, immersion and air. Watt Poultry USA. 12:18-24.
- McMeekin, T. A., and C. J. Thomas. 1978. Retention of bacteria on chicken skin after immersion in bacterial suspensions. J. Appl. Bacteriol. 45(3):383-387.
- Mead, G. C., V. M. Allen, C. H. Burton, and J. E. L. Corry. 2000. Microbial cross contamination during air chilling of poultry. Br. Poult. Sci. 41(2):158-162.
- Mead, G. C., W. R. Hudson, and M. H. Hinton. 1995. Effect of changes in processing to

- improve hygiene control on contamination of poultry carcasses with *Campylobacter*. *Epidemiol. Infect.* 115(3):495-500.
- Mead, P. S., L. Slutsker, V. Dietz, L. F. McCaig, J. S. Bresee, C. Shapiro, P. M. Griffin and R. V. Tauxe. 1999. Food-related illness and death in the United States. *Emerg. Infect. Dis.* 5(5):607-625.
- Mehyar, G. F., G. Blank, J. H. Han, A. Hydamaka, and R. A. Holley. 2005. Effectiveness of trisodium phosphate, lactic acid and commercial antimicrobials against pathogenic bacteria on chicken skin. *Food Prot. Trends.* 25:351-362.
- Meinersmann, R. J., W. E. Rigsby, N. J. Stern, L. C. Kelley, J. E. Hill, and M. P. Doyle. 1991. Comparative study of colonizing and noncolonizing *Campylobacter jejuni*. *Am. J. Vet. Res.* 52(9):1518-1522.
- MLG 41.04 United States Department of Agriculture. 2016. Laboratory Guidebook. Isolation and identification of *Campylobacter jejuni/coli/lari* from poultry rinse, sponge, and raw product samples. <http://www.fsis.usda.gov/>.
- Moore, J. E., M. D. Barton, I. S. Blair, D. Corcoran, J. S. Dooley, S. Fanning, I. Kempf, A. J. Lastovica, C. J. Lowery, M. Matsuda, D. A. McDowell, A. McMahon, B. C. Millar, J. R. Rao, P. J. Rooney, B. S. Seal, W. J. Snelling, and O. Tolba. 2006. The epidemiology of antibiotic resistance in *Campylobacter*. *Microbes Infect.* 8(7):1955-1966.
- Moore, J. E., M. Crowe, N. Heaney, and E. Crothers. 2001. Antibiotic resistance in *Campylobacter* spp. isolated from human feces (1980-2000) and foods (1997-2000) in Northern Ireland: an update. *J. Antimicrob. Chemother.* 48(3):455-457.
- Moore, J. E., T. S. Wilson, D. R. Wareing, T. J. Humphrey, and P. G. Murphy. 2002.

- Prevalence of thermophilic *Campylobacter* spp. in ready-to-eat foods and raw poultry in Northern Ireland. *J. Food Prot.* 65(8):1326-1328.
- Moran, A. P., and M. E. Upton. 1986. A comparative study of the rod and coccoid forms of *Campylobacter jejuni* ATCC 29428. *J. Appl. Bacteriol.* 60(2):103-110.
- Mulder, R. W. A. W., L. W. J. Dorresteyn, and J. van der Broek. 1978. Cross contamination during the scalding and plucking of broilers. *Br. Poult. Sci.* 19:61-70.
- Mulder, R. W. A. W., M. C. van der Hulst, and N. M. Bolder. 1987. *Salmonella* decontamination of broiler carcasses with lactic acid, l-cysteine, and hydrogen peroxide. *Poult. Sci.* 66:1555-1557.
- Murphy, C., C. Carroll, and K. N. Jordan. 2003. Induction of an adaptive tolerance response in the foodborne pathogen, *Campylobacter jejuni*. *FEMS Microbiol. Lett.* 223(1):89-93.
- Murphy, C., C. Carroll, and K. N. Jordan. 2006. Environmental survival mechanisms of the foodborne pathogen *Campylobacter jejuni*. *J. Appl. Microbiol.* 100(4):623-632.
- Nachamkin, I. 1995. *Campylobacter* and *Arcobacter*. Pages 483-491 in *Manual of Clinical Microbiology*, 6th ed. P. R. Murray, E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. J. Tenover, eds. Washington, D.C.
- Nachamkin, I., B. M. Allos, and T. Ho. 1998. *Campylobacter* species and Guillain-Barre syndrome. *Clin. Microbiol. Rev.* 11(3):555-567.
- Nadeau, E., S. Messier, and S. Quessy. 2002. Prevalence and comparison of genetic

- profiles of *Campylobacter* strains isolated from poultry and sporadic cases of campylobacteriosis in humans. *J. Food Prot.* 65(1):73-78.
- Nagel, G. M., L. J. Bauermeister, C. L. Bratcher, M. Singh, and S. R. McKee. 2013. *Salmonella* and *Campylobacter* reduction and quality characteristics of poultry carcasses treated with various antimicrobials in a post-chill immersion tank. *Int. J. Food Microbiol.* 165(3):281-286.
- Nauta, M. J., A. Hill, H. Rosenquist, S. Brynestad, A. Fetsch, P. van der Logt, A. Fazil, B. B. Christensen, E. Katsma, B. Borck, and A. H. Havelaar. 2009. A comparison of risk assessments on *Campylobacter* in broiler meat. *Int. J. Food Microbiol.* 129:107-123.
- National Antimicrobial Resistance Monitoring System (NARMS). 2015. NARMS integrated report: 2012-2013. Food and Drug Administration.
- National Chicken Council (NCC). 2018. Broiler chicken industry key facts 2018. <https://www.nationalchickencouncil.org/about-the-industry/statistics/broiler-chicken-industry-key-facts/>.
- National Institute of Allergy and Infectious Diseases (NIH). 2015. Campylobacteriosis. United States Department of Health and Human Services. <https://www.niaid.nih.gov/topics/campylobacteriosis/Pages/default.aspx>.
- Nelson, J. M., T. M. Chiller, J. H. Powers, and F. J. Angulo. 2007. Fluoroquinolone resistant *Campylobacter* species and the withdrawal of fluoroquinolones from use in poultry: a public health success story. *Clin. Infect. Dis.* 44(7):977-980.
- Newell, D. G., and H. C. Davison. 2003. *Campylobacter* - control and prevention.

- Page 470 in Chapter 22: Microbial Food Safety in Animal Agriculture. M. E. Torrence, and R. E. Isaacson, eds. Wiley-Blackwell Publishing.
- Newell, D. G., and J. A. Wagenaar. 2000. Poultry infections and their control at the farm level. Pages 497-510 in *Campylobacter*, 2nd ed. I. Nachamkin, M. J. Blaser, eds. Washington, D.C.
- Noormohamed, A., and M. K. Fakur. 2012. Incidence and antimicrobial resistance profiling of *Campylobacter* in retail chicken livers and gizzards. *Foodborne Pathog. Dis.* 9:617-624.
- Northcutt, J. K., M. E. Berrang, J. A. Dickens, D. L. Fletcher, and N. A. Cox. 2003a. Effect of broiler age, feed withdrawal, and transportation on levels of coliforms, *Campylobacter*, *Escherichia coli* and *Salmonella* on carcasses before and after immersion chilling. *Poult. Sci.* 82(1):169-173.
- Northcutt, J. K., R. J. Buhr, M. E. Berrang, and D. L. Fletcher. 2003b. Effects of replacement finisher feed and length of feed withdrawal on broiler carcass yield and bacteria recovery. *Poult. Sci.* 82(11):1820-1824.
- Northcutt, J. K., and S. M. Russell. 2010. General guidelines for implementation of HACCP in a poultry processing plant. The cooperative extension, University of Georgia. <http://www.caes.uga.edu/>.
- Northcutt, J. K., D. Smith, R. I. Huezos, and K. D. Ingram. 2008. Microbiology of broiler carcasses and chemistry of chiller water as affected by water reuse. *Poult. Sci.* 87(7):1458-1463.
- Northcutt, J. K., D. P. Smith, M. T. Musgrove, K. D. Ingram, and A. Hinton, Jr. 2005.

- Microbiological impact of spray washing broiler carcasses using different chlorine concentrations and water temperatures. *Poult. Sci.* 84(10):1648-1652.
- Oyarzabal, O. A. 2005. Reduction of *Campylobacter* spp. by commercial antimicrobials applied during the processing of broiler chickens: a review from the United States perspective. *J. Food Prot.* 68(8):1752-1760.
- Oyarzabal, O. A., C. Hawk, S. F. Bilgili, C. C. Warf, and G. K. Kemp. 2004. Effects of post-chill application of acidified sodium chlorite to control *Campylobacter* spp. and *Escherichia coli* on commercial broiler carcasses. *J. Food Prot.* 67(10):2288-2291.
- Pacholewicz, E., A. Swart, J. A. Wagenaar, L. J. Lipman, and A. H. Havelaar. 2016. Explanatory variables associated with *Campylobacter* and *Escherichia coli* concentrations on broiler chicken carcasses during processing in two slaughterhouses. *J. Food Prot.* 79(12):2038-2047.
- Padungtod, P., R. Hanson, D. L. Wilson, J. Bell, J. E. Linz, and J. B. Kaneene. 2002. Identification of *Campylobacter jejuni* isolates from cloacal and carcass swabs in chickens in Thailand by a 5' nuclease fluorogenic polymerase chain reaction assay. *J. Food Prot.* 65(11):1712-1716.
- Painter, J. A., R. M. Hoekstra, T. Ayers, R. V. Tauxe, C. R. Braden, F. J. Angulo, and P. M. Griffin. 2013. Attribution of foodborne illness, hospitalizations, and deaths to food commodities by using outbreak data, United States, 1998-2008. *Emerg. Infect. Dis.* 19(3):407-415.
- Park, S. F. 2002. The physiology of *Campylobacter* species and its relevance to their role as foodborne pathogens. *Int. J. Food Microbiol.* 74(3):177-188.

- Parveen, S., M. Taabodi, J. G. Schwarz, T. P. Oscar, J. Harter-Dennis, and D. G. White. 2007. Prevalence and antimicrobial resistance of *Salmonella* recovered from processed poultry. *J. Food Prot.* 70(11):2466-2472.
- Pearson, A. D., M. Greenwood, T. D. Healing, D. M. Rollins, M. Shahamat, J. Donaldson, and R. R. Colwell. 1993. Colonization of broiler chickens by waterborne *Campylobacter jejuni*. *Appl. Environ. Microbiol.* 59(4):987-996.
- Petracci, M., and C. Cavani. 2012. Muscle growth and poultry meat quality issues. *Nutrients.* 4(1):1-12.
- Petracci, M., S. Mudalal, E. Babini, and C. Cavani. 2014. Effect of white striping on chemical composition and nutritional value of chicken breast meat. *Ital. J. Anim. Sci.* 13(1):179-183.
- Petrak T., Z. Kalodera, P. Novakovic, and L. Gumhalter Karolyi. 1999. Bacteriological comparison of parallel and counter flow water chilling of poultry meat. *Meat Sci.* 53:269-271.
- PEW. 2016. Emerging pathogens in meat and poultry. Pages 1-73. PEW Charitable Trusts.
- Piddock, L. J. 1999. Mechanisms of fluoroquinolone resistance: an update 1994-1998. *Drugs.* 58(2):11-18.
- Rajkovic, A., N. Tomic, N. Smigic, M. Uyttendaele, P. Ragaert, and F. Devlieghere. 2010. Survival of *Campylobacter jejuni* on raw chicken legs packed in high-oxygen or high carbon dioxide atmosphere after the decontamination with lactic acid/sodium lactate buffer. *Int. J. Food Microbiol.* 140:201-206.
- Rasmussen, W. D. 1986. Lincoln's agricultural legacy. United States Department of

- Agriculture. <https://www.nal.usda.gov/lincolns-agricultural-legacy>.
- Rasschaert, G., V. Piessens, P. Scheldeman, S. Leleu, A. Stals, L. Herman, M. Heyndrickx, and W. Messens. 2013. Efficacy of electrolyzed oxidizing water and lactic acid on the reduction of *Campylobacter* on naturally contaminated broiler carcasses during processing. *Poult. Sci.* 92(4):1077-1084.
- Richardson, L. J., N. A. Cox, R. J. Buhr, and M. A. Harrison. 2011. Isolation of *Campylobacter* from circulating blood of commercial broilers. *Avian Dis.* 55(3):375-378.
- Ricke, S. C. 2014. Application of molecular approaches for understanding foodborne *Salmonella* establishment in poultry production. Pages 1-25 in *Advances in Biology*. Hindawi Publishing Corporation. Wiley and Sons, Inc.
- Ricke, S. C., E. J. van Loo, M. G. Johnson, and C. A. O'Bryan. 2012. Alternatives to traditional antimicrobials for organically processed meat and poultry. Pages 211-216 in *Organic Meat Production and Processing*.
- Ridley, A. M., V. M. Allen, M. Sharma, J. A. Harris, and D. G. Newell. 2008. Real-time PCR approach for detection of environmental sources of *Campylobacter* strains colonizing broiler flocks. *Appl. Environ. Microbiol.* 74(8):2492-2504.
- Riedel, C. T., L. Brondsted, H. Rosenquist, S. N. Haxgart, and B. B. Christensen. 2009. Chemical decontamination of *Campylobacter jejuni* on chicken skin and meat. *J. Food Prot.* 72(6):1173-1180.
- Rio, E., M. Panizo-Moran, M. Prieto, C. Alonso-Calleja, and R. Capita. 2007. Effect of various chemical decontamination treatments on natural microflora and sensory characteristics of poultry. *Int. J. Food Microbiol.* 115:268-280.

- Robinson, D. A. 1981. Infective dose of *Campylobacter jejuni* in milk. Br. Med. J. 282:1584.
- Rollins, D. M., and R. R. Colwell. 1986. Viable but nonculturable stage of *Campylobacter jejuni* and its role in survival in the natural aquatic environment. Appl. Environ. Microbiol. 52(3):531-538.
- Rosenquist, H., L. Boysen, C. Galliano, S. Nordentoft, S. Ethelberg, and B. Borck. 2009. Danish strategies to control *Campylobacter* in broilers and broiler meat: facts and effects. Epidemiol. Infect. 137(12):1742-1750.
- Rouger, A., O. Tresse, and M. Zagorec. 2017. Bacterial contaminants of poultry meat: sources, species, and dynamics. Microorganisms. 5(5):1-16.
- Russell, S. M. 2007. *Salmonella* reduction at scalding and picking: a big opportunity. <http://www.wattpoultryusa-digital.com/PoultryUSA/Article.aspx?io=12190>.
- Russell, S. M. 2012a. Verification of the efficacy of intervention strategies. Pages 243 – 257 in Controlling *Salmonella* in poultry production and processing. Boca Raton. CRC Press. London, N.Y.
- Russell, S. M. 2012b. *Salmonella* intervention strategies and testing methods differ greatly between the United States and Europe. Pages 259-267 in Controlling *Salmonella* in poultry production and processing. Boca Raton. CRC Press. London, N.Y.
- Russell, S. M. 2012c. Intervention strategies for reducing *Salmonella* prevalence on ready-to-cook chicken. Cooperative Extension. Bulletin 1222:1-13.
- Russell, S. M., D. L. Fletcher, and N. A. Cox. 1996. Spoilage bacteria of fresh broiler chicken carcasses. Poult. Sci. 74(12):2041-2047.

- Saenz, Y., M. Zarazaga, M. Lantero, M. J. Gastanares, F. Baquero, and C. Torres. 2000. Antibiotic resistance in *Campylobacter* strains isolated from animals, foods, and humans in Spain in 1997-1998. *Antimicrob. Agents Chemother.* 44(2):257-271.
- Saleha, A. A., G. C. Mead, A. L. Mead, and A. L. Ibrahim. 1998. *Campylobacter jejuni* in poultry production and processing in relation to public health. *J. World Poult. Sci.* 54(1):49-58.
- Sampathkumar, B., G. G. Khachatourians, and D. R. Korber. 2004. Treatment of *Salmonella enterica* serovar Enteritidis with a sublethal concentration of trisodium phosphate or alkaline pH induces thermotolerance. *Appl. Environ. Microbiol.* 70(8):4613-4620.
- Sams, A. R. 2001. First processing: slaughter through chilling. Pages 19-34 in *Poultry Meat Processing*. A. R. Sams, ed. CRC Press. Boca Raton, FL.
- Sams, A. R., and C. Z. Alvarado. 2010. Introduction to poultry meat processing. Pages 1-3 in *Poultry Meat Processing*. C. M. Owens, C. Z. Alvarado, and A. R. Sams, eds. CRC Press. Boca Raton, FL.
- Sanchez, M. X., W. M. Fluckey, M. M. Brashears, and S. R. McKee. 2002. Microbial profile and antibiotic susceptibility of *Campylobacter* spp. and *Salmonella* spp. in broilers processed in air chilled and immersion chilled environments. *J. Food Prot.* 65(6):948-956.
- Scallan, E., P. M. Griffin, F. J. Angulo, R. V. Tauxe, and R. M. Hoekstra. 2011a. Foodborne illness acquired in the United States - unspecified agents. *Emerg. Infect. Dis.* 17(1):16-22.
- Scallan, E., R. M. Hoekstra, F. J. Angulo, R. V. Tauxe, M. Widdowson, S. L. Roy, J. L.

- Jones, and P. M. Griffin. 2011b. Foodborne illness acquired in the United States - major pathogens. *Emerg. Infect. Dis.* 17(1):7-15.
- Schreurs, F. J. G. 2000. Post mortem changes in chicken muscle. *World's Poult. Sci. J.* 56:319-346.
- Scientific Committee on Veterinary Measures Relating to Public Health (SCVMRPH). 1998. Benefits and limitations of antimicrobial treatments for poultry carcasses. European Commission. Brussels, Belgium. https://ec.europa.eu/food/sites/food/files/safety/docs/sci-com_scv_out14_en.pdf.
- Sellars, M. J., S. J. Hall, and D. J. Kelly. 2002. Growth of *Campylobacter jejuni* supported by respiration of fumarate, nitrate, nitrite, trimethylamine-*N*-oxide, or dimethyl sulfoxide requires oxygen. *J. Bacteriol.* 184(15):4187-4196.
- Shakil, S., R. Khan, R. Zarrilli, and A. U. Khan. 2008. Aminoglycosides versus bacteria-a description of the action, resistance mechanism, and nosocomial battleground. *J. Biomed. Sci.* 15(1):5-14.
- Skirrow, M. B. 1977. *Campylobacter* enteritis: a new disease. *Br. Med. J.* 2:9-11.
- Simaluiza, R. J., Z. Toledo, S. Ochoa, and H. Fernandez. 2015. The prevalence and antimicrobial resistance of *Campylobacter jejuni* and *Campylobacter coli* in chicken livers used for human consumption in Ecuador. *J. Anim. Vet. Adv.* 14:6-9.
- Singh, M. 2013. Intervention strategies for control of *Campylobacter* during poultry processing. Department of Poultry Science. Auburn University. https://confexrecordings.s3.amazonaws.com/asas/RM13/paper471_1.pdf.
- Smith, R. N., R. N. Andersen, and P. E. Kolenbrander. 1991. Inhibition of intergeneric

- coaggregation among oral bacteria by cetylpyridinium chloride, chlorhexidine digluconate and octenidine dihydrochloride. *J. Periodontal Res.* 26(5):422-428.
- Smith, K. E., J. M. Besser, C. W. Hedberg, F. T. Leano, J. B. Bender, J. H. Wicklund, B. P. Johnson, K. A. Moore, and M. T. Osterholm. 1999. Quinolone-resistant *Campylobacter jejuni* infections in Minnesota, 1992-1998. *N. Engl. J. Med.* 340(20):1525-1532.
- Smith, J., S. Corkran, S. R. McKee, S. F. Bilgili, and M. Singh. 2015. Evaluation of post chill applications of antimicrobials against *Campylobacter jejuni* on poultry carcasses. *J. Appl. Poult. Res.* 24:451-456.
- Smulders, F. 1995. Preservation by microbial decontamination; the surface treatment of meats by organic acids. In *Print: New Methods of Food Preservation*. G. W. Gould, ed. Blackie Academic and Professional, London.
- Sofos, J. N., G. Flick, G. Nychas, C. A. O'Bryan, S. C. Ricke, and P. G. Crandall. 2013. Meat, Poultry and Seafood. Pages 111-167 in *Food Microbiology: Fundamentals and Frontiers*. 4th ed. M. P. Doyle, and R. L. Buchanan, eds. ASM Press. Washington, D.C.
- Statistica. 2013. StatSoft, Inc. Version 12.0, ed. Tulsa, OK, USA.
- Stopforth, J. D., R. O'Connor, M. Lopes, B. Kottapalli, W. E. Hill, and M. Samadpour. 2007. Validation of individual and multiple-sequential interventions for reduction of microbial populations during processing of poultry carcasses and parts. *J. Food Prot.* 70(6):1393-1401.
- Stern, N. J., B. Wojton and K. Kwiatek. 1992. A differential selective medium and dry ice generated atmosphere for recovery of *Campylobacter jejuni*. *J. Food Prot.*

55:514-517.

- Stopforth, J. D., R. O'Connor, M. Lopes, B. Kottapalli, W. E. Hill, and M. Samadpour. 2007. Validation of individual and multiple-sequential interventions for reduction of microbial populations during processing of poultry carcasses and parts. *J. Food Prot.* 70:1393-1401.
- Strachan, N. J. C., M. MacRae, A. Thomson, O. Rotariu, I. D. Ogden, and K. J. Forbes. 2012. Source attribution, prevalence and enumeration of *Campylobacter* spp. from retail liver. *Int. J. Food Microbiol.* 153:234-236.
- Sukted, N., P. Tuitemwong, K. Tuitemwong, W. Poonlapdecha, and L. E. Erickson. 2017. Inactivation of *Campylobacter* during immersion chilling of chicken carcasses. *J. Food Eng.* 202:25-33.
- Talsma, E., W. G. Goettsch, H. L. Nieste, P. M. Schrijnemakers, and M. J. Sprenger. 1999. Resistance in *Campylobacter* species: increased resistance to fluoroquinolones and seasonal variation. *Clin. Infect. Dis.* 29(4):845-848.
- Tamblyn, K. C., and D. E. Conner. 1997. Bactericidal activity of organic acids against *Salmonella* Typhimurium attached to broiler chicken skin. *J. Food Prot.* 60(6):623-633.
- Tauxe, R. V. 1992. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. Pages 9-19 in *Campylobacter jejuni: current status and future trends*. I. Nachamkin, M. J. Blaser, and L. S. Tompkins, eds. American Society for Microbiology. Washington, D.C.
- Taylor, J. B., and D. W. Wohlers. 2004. Toxicological profile for chlorine dioxide and

chlorite. U.S. Department of Health and Human Services.

<http://www.atsdr.cdc.gov/>.

Thomas, C. J., and T. A. McMeekin. 1980. Contamination of broiler carcass skin during commercial processing procedures: an electron microscopic study. *Appl. Environ. Microbiol.* 40(1):133-144.

Thomas, C. J., and T. A. McMeekin. 1982. Effect of water immersion on the microtopography of the skin of chicken carcasses. *J. Sci. Food Agr.* 33:549-554.

Thompson, T. M., M. E. Berrang, N. A. Cox, and R. J. Meinersmann. 2018.

Campylobacter prevalence in retail chicken liver. *Poult. Sci.* (Suppl 1).

Travers, K., and M. Barza. 2002. Morbidity of infections caused by antimicrobial resistant bacteria. *Clin. Infect. Dis.* 34(Suppl. 3):S131-S134.

United States Department of Agriculture (USDA). 1984. Regulations under the poultry products inspection act. Regulations Office, Policy and Program Planning, Food Safety and Inspection Service. U.S. Department of Agriculture. Washington, D.C.

United States Department of Agriculture (USDA). 2001. Retained water in raw meat and poultry products. Poultry chilling requirements. Final Rule. 9 CFR Parts 381 and 441. 66:1750-1772.

United States Department of Agriculture (USDA). 2002. The use of chlorine dioxide as an antimicrobial agent in poultry processing in the United States. USDA-FSIS, Office of International Affairs. November 2002.

United States Department of Agriculture (USDA). 2016a. Overview of the United States slaughter industry. <https://usda.mannlib.cornell.edu>

- United States Department of Agriculture (USDA). 2016b. USDA finalizes new food safety measures to reduce *Salmonella* and *Campylobacter* in poultry. <http://www.usda.gov/wps/portal/usda/>.
- United States Department of Agriculture (USDA). 2017a. Poultry production and value summary. National Agricultural Statistics Service. <https://mannlib.cornell.edu>.
- United States Department of Agriculture (USDA). 2017b. Cost estimates of foodborne illnesses. <https://www.ers.usda.gov/data-products/cost-estimates-of-foodborne-illnesses/>.
- United States Department of Agriculture (USDA). 2018. About the U.S. Department of Agriculture. <https://www.usda.gov/our-agency/about-usda>.
- USDA-FSIS. 1996. Pathogen reduction: hazard analysis and critical control point (HACCP) systems, final rule. Fed. Regist. 61:38806.
- USDA-FSIS. 2008. Compliance guidelines for controlling *Salmonella* and *Campylobacter* in poultry. 2nd edition. <http://www.fsis.usda.gov/>.
- USDA-FSIS. 2014. Chicken from Farm to Table. U.S. Department of Agriculture, Food Safety and Inspection Service. <https://fsis.usda.gov/>.
- USDA-FSIS. 2015. Public health effects of raw chicken parts and comminuted chicken and turkey performance standards. <http://www.fsis.usda.gov/shared/PDF/Effects-Performance-Standards-Chicken-Parts-Comminuted.pdf>.
- USDA-FSIS. 2016a. Health hazard information sheet: peroxyacetic acid (PAA). FSIS Environmental, Safety and Health Group (ESHG). <http://www.fsis.usda.gov/>.
- USDA-FSIS. 2016b. Isolation and identification of *Campylobacter jejuni/coli/lari* from

poultry rinse, sponge, and raw product samples. Laboratory Guidebook MLG 41.04.

USDA-FSIS. 2016c. Sanitation performance standards compliance guide. Title 9. Code of Federal Regulations 416.

USDA-FSIS. 2018. Table of safe and suitable ingredients October 2018. https://www.fsis.usda.gov/wps/wcm/connect/ce40e7ae-3d55-419e-9c68-a1b6fefcd4de/7120.1_Table_2.pdf?MOD=AJPERES.

Van de Giessen, A. W., B. P. Bloemberg, W. S. Ritmeester, and J. J. Tilburg. 1996. Epidemiological study on risk factors and risk reducing measures for *Campylobacter* infections in Dutch broiler flocks. *Epidemiol. Infect.* 117:245-250.

Vandamme, P. 2000. Taxonomy of the family Campylobacteraceae. Pages 3-26 in *Campylobacter*. I. Nachamkin, and M. J. Blaser, eds. American Society for Microbiology. Washington, D.C.

Vashin, I., T. Stoyanchev, C. Ring, and V. Atanassova. 2009. Prevalence of *Campylobacter* spp. in frozen poultry giblets at Bulgarian retail markets. *Trakia J. Sci.* 7:55-57.

Vasseur, C., L. Baverel, M. Hebraud, and J. Labadie. 1999. Effect of osmotic, alkaline, acid, or thermal stress on the growth and inhibition of *Listeria monocytogenes*. *J. Appl. Microbiol.* 86:469-476.

Veluz, G. A., S. Pitchiah, and C. Z. Alvarado. 2012. Attachment of *Salmonella* serovars and *Listeria monocytogenes* to stainless steel and plastic conveyor belts. *Poult. Sci.* 91(8):2004-2010.

- Vugia, D., A. Cronquist, J. Hadler, P. Blake, D. Blythe, K. Smith, D. Morse, P. Cieslak, T. Jones, D. Goldman, J. Guzewich, F. Angulo, P. Griffin, R. Tauxe, and K. Kretsinger. 2003. Preliminary FoodNet data on the incidence of infection with pathogens transmitted commonly through food - selected sites, United States. *Morb. Mortal Wkly Rep.* 53(16):338-343.
- Waldroup, A., C. Coleman, and K. Beers. 2000. Commercialization of cetylpyridinium chloride for use in the poultry industry. U.S. Food Additive Petition No. 2A4736, Appendix VII. Food and Drug Administration, Department of Health and Human Services. Washington, D.C.
- Wang, Y., C. Lehane, K. Ghebremeskel, and M. A. Crawford. 2010. Modern organic and broiler chickens sold for human consumption provide more energy from fat than protein. *Public Health Nutr.* 13(3):400-408.
- Wempe, J. M., C. A. Genigeorgis, T. B. Farver, and H. I. Yusufu. 1983. Prevalence of *Campylobacter jejuni* in two California chicken processing plants. *Appl. Environ. Microbiol.* 45(2):355-359.
- Whyte, P., J. D. Collins, K. McGill, and C. Monahan. 2007. Assessment of sodium dichloroisocyanurate in the control of microbiological cross-contamination in broiler carcass immersion chilling systems. *J. Food Safety.* 22(1):55-65.
- Whyte, R., J. A. Hudson, and C. Graham. 2006. *Campylobacter* in chicken livers and their destruction by pan frying. *Lett. Appl. Microbiol.* 43:591-595.
- Wideman, N., M. Bailey, S. F. Bilgili, H. Thippareddi, L. Wang, C. Bratcher, M. Sanchez-Plata, and M. Singh. 2016. Evaluating best practices for *Campylobacter* and *Salmonella* reduction in poultry processing plants. *Poult. Sci.* 95(2):306-315.

- Williams, A., and O. A. Oyarzabal. 2012. Prevalence of *Campylobacter* spp. in skinless, boneless, retail broiler meat from 2005 through 2011 in Alabama, USA. *BMC Microbiol.* 12:184.
- Willis, W. L., and C. Murray. 1997. *Campylobacter jejuni* seasonal recovery observations of retail market broilers. *Poult. Sci.* 76(2):314-317.
- Zamora-Sanabria, R., and A. M. Alvarado. 2017. Preharvest *Salmonella* risk contamination and the control strategies. Intech: Open Science: Chapter 11.
- Zeitoun, A., F. Devlieghere, and J. Debevere. 1996. Increasing the shelf-life of poultry meat using acid decontamination and modified atmosphere packaging. Pages 215-225 in *Microbial Methods for the Meat Industry, Concerted Action CT94-1456*. M. H., Hinton, and C. Rowlinson, eds. University of Bristol Press. Bristol.
- Zhao, T., and M. P. Doyle. 2006. Reduction of *Campylobacter jejuni* on chicken wings by chemical treatments. *J. Food Prot.* 69(4):762-767.
- Zhao, C., B. Ge, J. D. Villena, R. Sudler, E. Yeh, S. Zhao, D. G. White, D. Wagner, and J. Meng. 2001. Prevalence of *Campylobacter* spp., *Escherichia coli*, and *Salmonella* serovars in retail chicken, turkey, pork, and beef from the greater Washington, D.C. Area. *Appl. Environ. Microbiol.* 67(12):5431-5436.
- Zweifel, C., and R. Stephan. 2012. Microbial decontamination of poultry carcasses. Pages 60-95 in *Microbial Decontamination in the Food Industry*. A. Demirci, and M. O. Ngadi, eds. Woodhead Publishing, Cambridge, UK.