Best Management Practices for *Salmonella* and *Campylobacter* Control in Poultry Processing Plants

by

Nathan Edwin Wideman

A thesis submitted to the Graduate Faculty of
Auburn University
in partial fulfillment of the
requirements for the Degree of
Master of Science

Auburn, Alabama
December 14, 2013

Keywords: *Campylobacter*, *Salmonella*, foodborne illness

Approved by

Manpreet Singh, Chair, Associate Professor, Department of Poultry Science
Sacit Bilgili, Professor, Department of Poultry Science
Christy Bratcher, Associate Professor, Department of Animal Sciences
Luxin Wang, Assistant Professor, Department of Animal Sciences

Abstract

Campylobacter and *Salmonella* are the top two pathogens associated with poultry-related foodborne illness, currently responsible for over 800,000 illnesses annually (*13*). The reduction of these pathogens on commercial poultry products at the processing plant level is crucial to reducing the incidence of foodborne illness, as significant reduction in these pathogens prior to reaching the processing plant may not be easy. Currently, poultry processing plants use multiple intervention methods at various sites to reduce the *Campylobacter* and *Salmonella* load as much as possible, with few plant management practices being standard throughout poultry processing. The determination of these best practices is necessary to maximize the reduction of these foodborne pathogens while minimizing the cost and waste within the processing plant.

Six poultry processing plants were analyzed in the Southeastern United States in order to evaluate their current pathogen control practices, suggest changes, and evaluate to the effectiveness of the changes. Surveys were sent to the plant Quality Assurance managers to determine production levels, antimicrobial interventions, and current pathogen testing practices. Then an initial sampling set was taken at each plant, at sites that included carcass samples before any pre-evisceration intervention, after exiting the inside-outside bird washer (IOBW), after exiting the pre-chiller, after exiting the primary chiller, and after exiting any post-chill intervention, as well as a water sample from each scalder, pre-chiller, primary chiller, and

post-chill dip tank or finishing chiller, and finally a pooled manure sample to analyze incoming microbial load. Enumerations were performed for *Campylobacter* and *Salmonella* as well as enrichments for *Campylobacter* and *Salmonella*. After the initial sample set, each plant was suggested several changes to be made and once the changes were implemented a second sampling set was conducted to determine the effectiveness of these changes.

The analysis of the various practices showed that peracetic acid (PAA) was the most effective antimicrobial currently in use. The use of a post-chill antimicrobial immersion tank and/or use of Cetylpyridinium Chloride CPC spray cabinet also displayed a further reduction in microbial levels when the primary chiller was not sufficient. Slight microbial buildup occurred in the immersion tanks, however effective cleaning techniques and chiller maintenance may minimize these negative effects. Further research on the use of PAA, CPC, and post-chill immersion tanks may help further optimize plant pathogen control practices throughout the United States.

Acknowledgments

I would like to thank my major advisor Dr. Manpreet Singh for his confidence, help, support, and motivation over the past two years. I would also like to thank Dr. Manpreet Singh for his initial willingness to take a chance and allow me to prove myself as a graduate student. I would like to thank Dr. Sacit Bilgili, Dr. Christy Bratcher, and Dr. Luxin Wang for their review and assistance in my research. I am grateful for my family and friends for the love, support, and encouragement that they continue to give.

Table of Contents

List of Tables

List of Figures

List of Abbreviations

ASC	Acidified sodium chlorite
BPW	Buffered Peptone Water
CFU	Colony-Forming Unit
CPC	Cetylpyridinium chloride
HACCP	Hazard Analysis and Critical Control Points
IOBW	Inside-Outside Bird Washer
OLR	On-line Reprocessing
PPM	Parts per million
PAA	Peracetic acid
PAHP	Peracetic acid and hydrogen peroxide
TSP	Trisodium phosphaste
USDA	United States Department of Agriculture

Introduction

Campylobacter species are Gram-negative, motile, spiral-rod bacteria that are part of the natural gut microflora of poultry as well as other food-producing animals. Similarly, *Salmonella* species are Gram-negative, motile, rod bacteria found on many food sources, including in the natural gut microflora of poultry. The consumption of *Campylobacter*- and/or *Salmonella*-contaminated products can lead to Campylobacteriosis and Salmonellosis infections respectively. The symptoms of these infections usually include nausea, vomiting, abdominal cramps, diarrhea, fever, and headache. In severe cases, further complications can occur including Guillain-Barré syndrome as a result of *Campylobacter* infection and Reiter's Syndrome as a result of *Salmonella* infection.

The CDC estimates that over 1.8 million cases of foodborne illness were caused by consumption of *Salmonella*- and *Campylobacter*-contaminated food products (*5*). FoodNet also found that the incidence rate of *Campylobacter* and *Salmonella* infections had increased by 14% and 6% respectively from 2006 to 2011. The low infectious dose of the bacteria (i.e., as low as a single cell for *Salmonella* infection and 500 cells for *Campylobacter* infection) along with the large number of food sources that have been discovered to harbor these bacteria make the reduction and elimination of these infectious organisms a high priority for the food industry.

As a result of recent outbreaks of *Salmonella* from previously-assumed safe food sources, as well as a continuing increase in the incidence of foodborne illness in the United States, the Food Safety Modernization Act (FSMA) was signed into law in 2011. This law shifted the food safety focus from a reaction to foodborne outbreaks to the prevention of the outbreaks. With this law enacted, the Food and Drug Administration (FDA) would be able to enforce stricter regulations and performance standards in the food industry, as well as build a national food safety system so that communication and management of problems could be as efficient as possible.

When investigating the foodborne sources of *Campylobacter* and *Salmonella* infection, the Emerging Pathogens Institute determined in 2011 that *Campylobacter*-contaminated poultry and *Salmonella*-contaminated poultry were the #1 and #4 food-pathogen pairs as a cause of foodborne illness in the United States. As a result of this they determined that contaminated poultry had the greatest public health impact of any food currently in production, resulting in over 1.5 million infections annually and $2.4 billion in costs of illness annually (*13*).

To combat the high risk of poultry-related foodborne illness, new *Campylobacter* and *Salmonella* performance standards were issued in 2011 by the United States Department of Agriculture Food Safety and Inspection Service (USDA-FSIS). These microbial performance standards include maximum 5 *Salmonella*-positive samples out of a 51-sample set, and maximum of 8 *Campylobacter*-positive samples out of a 51-sample set. Mandatory retesting is required for plants that fail initial tests, and the identity of such plants is made publically

available (*7*). Meeting these new performance standards is a high priority for poultry processing plants, as continued failed tests could lead to harsh penalties and potential shutdown of the plant. However the reduction of *Salmonella* and *Campylobacter* in poultry production has not been easy.

The first issue is control of the spread of *Salmonella* and *Campylobacter* before the birds reach the processing plant. The new performance standards and stricter regulations focus on the processing plant level of production, and currently there are few incentives to reduce the contamination and spread of the bacteria on the farm. In addition, the ability of *Salmonella* and *Campylobacter* contamination to go undetected in the host bird and the quick spread of the pathogens within a flock make reducing the rate of contamination at the farm difficult. Therefore it is left up to the poultry processing plant to eliminate or reduce any pathogen on the birds along the processing line. This leads to the issue of determining the best pathogen reduction practices for reduction and elimination of *Campylobacter* and *Salmonella* in poultry processing. The physical processing steps are standard among most poultry processing plants. Counter-flow scalders are used to remove organic material from the birds as well as soften the feather follicles for easy feather removal. The birds are also subjected to several rinses to further remove any organic material from the carcass, including a rinse following evisceration in an inside-outside bird washer (IOBW). USDA regulation mandates that the birds must then be chilled to <40°F within 6 hours of slaughter, which is usually accomplished using a pre-chiller and a primary chiller.

Where poultry processing plants vary is in the intervention points and antimicrobials used to prevent *Salmonella* and *Campylobacter* contamination of poultry carcasses from reaching the end of the processing line. The incoming bacterial load and subsequent *Salmonella* and *Campylobacter* load can vary from bird to bird, and so intervention strategies must take into account this variation when analyzing overall effectiveness. In addition, intervention strategies must be able to reduce or eliminate higher incoming bacterial loads while preventing cross-contamination so that the processing plant can meet the stricter USDA-FSIS performance standards. The low infective dose of both pathogens make it paramount that plants meet the stricter standards as any positive sample is a potential risk for infection to the consumer.

There is currently much debate on the effectiveness of intervention strategies at several points in the processing line. The use of chemicals such as chlorine and citric acid in the scalders may reduce the bacterial load, however high levels of organic matter in the scalder water often reduce their effectiveness. The use of chlorine in IOBW is a standard practice in most plants, however the use of chlorine in the pre-chiller, primary chiller, and any post-chill intervention may not be as effective as other chemicals such as PAA and CPC. The use of a post-chill intervention such as a dip tank, finishing chiller, or a drip-bar system is the last intervention point to reduce post-chill *Salmonella*- or *Campylobacter*-positive carcasses from reaching the end of the processing line.

There are many variables to take into account when determining the effectiveness of a plant's current pathogen intervention methods. Current intervention strategies may only work due to the low initial bacterial load on incoming carcasses, and may be ineffective if the

bacterial load were to increase. Variation in antimicrobial use concentration, contact time, pH of the water, bird size, and total immersion versus surface spray alter the effectiveness of intervention strategies.

In order to run a processing plant as efficiently as possible, intervention strategies must be analyzed to determine if they are meeting the industry standards, if they are cost-effective, and if they could be improved. With *Campylobacter*- and *Salmonella*-contaminated poultry continuing to be a major source of foodborne illness, immediate updates and optimizations to current processing plant intervention practices are necessary to reduce the risk of illness and prevent future outbreaks.

Chapter 1: Literature Review

1. History of *Campylobacter*

Campylobacter strains are gram-negative bacilli that have corkscrew shape motility, which is characteristic and helpful in identification. However as the organisms age or are stressed the bacteria can become more cocci in shape and lose their corkscrew motility. They are also motile, propelled by unipolar or bipolar flagella. A temperature of 42°C (the internal temperature of poultry) and a microaerophillic (5% O_2, 15% CO_2, 80% N_2) environment are the ideal growing conditions for these organisms, however they can survive outside these conditions. Even in ideal conditions the organisms have a slow growth rate, usually requiring at least 48 hours for proper isolation and identification.

Campylobacter species were originally identified as a cause of septic abortions in cattle and sheep (*38*), being misidentified as *Vibrio fetus*. When these animal infections were compared to human *V. fetus* infections (causing diarrhea), the organisms appeared to be identical. In 1973, it was determined that the genus *Vibrio* did not correctly describe *V. fetus*' characteristics (*77*), and so *V. fetus*, along with *V. coli, V. jejuni, V. sputorum, and V. bubulus* were transferred to the new genus of *Campylobacter.* New selective media allowed for the isolation of *Campylobacter* strains from stool samples, and by the late 1980s, *Campylobacter* was determined to be one of the most common causes of bacterial gastrointestinal illness worldwide (*1*).

Campylobacter infection is the fourth most common causes of domestic foodborne illness, estimated to affect over 800,000 people annually (*5*). It is also the third most common cause of foodborne illness-related hospitalization and fifth most common cause of foodborne illness-related death. *Campylobacter jejuni* is the most common strain associated with infection, and at one point it was believed to be responsible for over 99% of infections in the United States (*26*). Incidents of *Campylobacter* infection are usually sporadic, and typically require an infective dose of 500 cells or more (*61*). Typical symptoms of *Campylobacter* infection, or Campylobacteriosis, include diarrhea, fever, and abdominal cramps. *Campylobacter* infections occur by consuming contaminated food or through fecal-oral means, and are usually self-limiting (lasting up to 1 week) with antibiotics rarely being used. When antibiotics are used, ciprofloxacin and azithromycin are the drugs of choice, as *Campylobacter* strains have become increasingly resistant to fluoroquinolones (*1*). In rare cases further complications can result from the infection. Guillain-Barré syndrome is a serious sequel resulting from *Campylobacter* infections (*2*), resulting in acute neuro-muscular paralysis. *Campylobacter* infections are also associated with Reiter's syndrome (*57*), causing reactive arthritis (pain particularly in the knee joint), eye irritation, and painful urination. Death from *Campylobacter* infections are rare but can occur with infants, the elderly and immunocompromised persons being most at risk.

To help lower the risk of *Campylobacter* infection as a result of poultry consumption, new performance standards were issued by the Food Safety and Inspection Service in 2011, allowing for only 8 positive samples out of a 51 sample set post-chill in poultry processing plants. *Campylobacter* testing will become a higher priority in processing plants, and processing

plants failed tests will be met with stricter penalties. Despite these new regulations *Campylobacter* contamination is still a top issue in the poultry industry (*7*).

2. History of *Salmonella*

Salmonella strains are gram-negative, bacilli that can be found typically in the intestinal tract of humans and other animals, although they can be found in numerous other food sources. *Salmonella* cells are motile, using peritrichous flagella for movement. When culturing *Salmonella,* ideal growth occurs over 24 hours in a 37°C, aerobic environment. However being facultative anaerobes they can easily colonize and survive in the intestinal tract of many different animals, and are able to survive in soil and aquatic environments.

Salmonella was first discovered in 1884 when *Salmonella choleraesuis* was isolated from pigs with hog cholera (*71*), and have long since been identified as a human pathogen. Currently, two species make up the genus *Salmonella* (*Salmonella bongori* and *Salmonella enterica*); however there are more than 2,500 serovars of *S. enterica.* Typical symptoms of *Salmonella* infection, or Salmonellosis, include diarrhea, fever, and abdominal cramps 1-3 days after infection. The infection is transmitted by food as well as fecal-oral contact, and is typically self-limiting, lasting up to a week. Although rare in developed nations, *S.* Typhi and *S.* Paratyphi can cause typhoid fever and enteric fever respectively, leading to systemic infections. In rare cases, long-term Salmonellosis can lead to Reiter's Syndrome, causing joint pain, eye irritation, and painful urination. Death from Salmonellosis is rare with infants, the elderly and immunocompromised individuals being most at risk.

In the 1980s, *S.* Enteritidis infection emerged as a major cause of foodborne illness. From 1976 to 1985, the rate of *S.* Enteritidis infection increased five-fold and total *Salmonella* infection rates increased almost three-fold (*4*). In 1990, *S.* Enteritidis surpassed *S.* Typhimurium as the top serotype reported isolated from *Salmonella* infections in the United States (*43*). From 1985 to 1999, 389 outbreaks of *S.* Enteritidis were reported, with 298 being associated with egg products (*53*). As a result of these and other outbreaks, sweeping reforms to egg regulations were applied by the USDA to lower future incidences of *S.* Enteritidis infection. In 1996, the Foodborne Disease Active Surveillance Network (FoodNet) was created in an effort to combine lab resources for the reporting and tracking of bacterial infections within the United States. In 1997, use of the Hazard Analysis and Critical Control Points (HACCP) system became mandatory in United States processing plants to help combat foodborne illnesses including Salmonellosis. In 1999, the President's Council on Food Safety announced the Egg Safety Action Plan, with the goal of reducing egg-associated *S.* Enteritidis illness by half by 2005, and eliminating egg-associated illness by 2010. Currently, *Salmonella* infections are responsible for an estimated 1 million foodborne illnesses a year (*5*). *Salmonella* infections are also the top cause of foodborne illness-related hospitalizations and foodborne illness-related deaths. In 2011, new *Salmonella* performance standards were implemented by the Food Safety and Inspection Service to further reduce the incidence of *Salmonella*-contaminated poultry. The number of *Salmonella*-positive samples in a 51 sample set has been reduced to 5 (down from 20%). Any processing plant that fails sampling will be quickly resampled, and the identity of the company made public. Stricter penalties may also be applied to plants that continue to fail performance standard testing (*7*).

3. Issues in Poultry

Current numbers show that an estimated 1.5 million people in the United States are infected with a poultry-related foodborne illness annually, resulting in over $2.4 billion in costs of illness and making poultry the top food type associated with foodborne illness (*13*). Of this, *Campylobacter*-contaminated poultry is responsible for over 600,000 illnesses, and *Salmonella*-contaminated poultry is responsible for over 220,000 illnesses. If *Salmonella*-contaminated eggs are included, the number of illnesses rises to 330,000 illnesses (*13*). Due to the lack of consumer reporting of foodborne-related illnesses, these numbers may be underestimated. The reduction of poultry-related foodborne illnesses is one of the highest priorities in foodborne illness prevention.

3.1 *Campylobacter*

Over the past decade, *Campylobacter*-contaminated poultry has remained steady in the number of illnesses reported. However, *Campylobacter* contamination of poultry flocks is still an important issue that requires multiple hurdles to reduce the risk of contamination. The initial contamination vector can vary depending on the poultry farm. Insects within the poultry houses such as flies and beetles are potential carriers (*64*, *31*). Rodents also can carry *Campylobacter* into broiler housing (*20*). However, the most common sources of *Campylobacter* contamination in broiler flocks are typically through the water supply and fecal-oral transmission. If the drinking water for the birds is unchlorinated, *Campylobacter* can easily survive (although it cannot grow) and enter the birds digestive tracts (*56*). Farm workers can bring in *Campylobacter* on their clothing and boots, depositing the organism in the litter, air and drinking water in the housing (*34*). After the organism is introduced into the broiler flock, it

is quickly spread. There is some evidence to suggest vertical transmission of *Campylobacter* between the hen and the chick (*55*) and colonization of the birds can also occur within a few weeks of hatching by the boots and clothing of farm employees (*28*).

Once *Campylobacter* has colonized the gut of a bird, the caprophagic nature of chickens facilitates the fecal-oral spread within the flock (*44*). The use of communal drinking water sources also allows for a rapid spread within flocks after initial *Campylobacter* introduction, with up to 100% infection occurring within a few days of initial colonization (*67*). The microaerophillic atmosphere within the gut and the body temperature of the broiler (42°C) are ideal conditions for *Campylobacte*r growth. *Campylobacter* colonizes throughout the gut of the broiler, however the highest numbers of *Campylobacter* are usually found in the mucosal layer of the caecum, sometimes reaching as high as 10^9 CFU/g of contents within 3 days of contamination (*46*). Since *Campylobacter* is a commensal organism in poultry, there are no visible symptoms of contamination. The commensal nature and rapid flock spread makes farm-level control of campylobacter difficult.

During transport and processing, cross-contamination is the main source of *Campylobacter* spread in broilers. Feed withdrawal must occur between 8-12 h prior to processing to reduce fecal cross-contamination as much as possible. If the feed is withdrawn less than 8 h before processing, the gut of the broiler may still contain a large amount of fecal matter, with release occurring during transport. If the feed is withdrawn greater than 12 h before processing, the intestinal tract becomes lose and weak. This causes the tract to easily tear during the evisceration process, causing fecal contamination of the bird during processing

as well as increasing the risk of cross-contamination with nearby birds. During transport, stress and confinement allows for fecal contamination to be spread between birds within each cage. In addition, cages are frequently unwashed/disinfected or not washed/disinfected to an adequate level between uses, allowing for further cross-contamination (*30*).

After transport, *Campylobacter* cross-contamination occurs in the scalders, pickers, chillers and post-chill dip tanks. Scalders usually operate at a temperature of 50°C to 55°C, which is not hot enough to eliminate *Campylobacter* from the water supply. Due to the high organic load, the use of an antimicrobial is impractical, and so the potential for cross-contamination between birds passing through the scalder tanks is high. *Campylobacter jejuni* has also been recovered from the feather pickers in processing plants (*80*). The rubber fingers of the pickers used to remove the feathers can easily pick up and spread *Campylobacter* from a contaminated bird, allowing for a high potential for cross-contamination on uncontaminated birds. Further studies have supported this hypothesis, showing significantly higher counts of *Campylobacter* after birds passed through the feather picker (*16*). In pre-chillers and primary chill tanks, low levels of *Campylobacter* may survive the antimicrobials added to the chiller water, potentially cross-contaminating the birds in the final processing steps. Even with current intervention methods, complete elimination of *Campylobacter* from all poultry carcasses is not expected. Retail chicken should not be assumed to be *Campylobacter*-free, and one study showed a rate of 98% contamination for retail chicken meat, albeit at low concentration. (*73*).

3.2 *Salmonella*

Despite the stricter poultry regulations, HACCP implementation, and FoodNet implementation as a result of the *S.* Enteritidis epidemic of the 1980's and 1990's, foodborne illnesses due to *Salmonella* infection has remained unchanged. Like *Campylobacter*, the elimination of *Salmonella* from poultry requires the focus on all steps in poultry processing.

There are many vectors in which *Salmonella* can be brought into the broiler housing and hence, can be difficult to determine the primary source of contamination. Rodents are an animal reservoir for *Salmonella,* acquiring the pathogens from outside sources and then spreading the pathogens throughout the housing through contaminated droppings (*24*). Insects such as flies and beetles are also potential carriers of *Salmonella* into the poultry housing (*49*, *25*), and can contaminate the water supply as well as the litter. Dust in the broiler housing can also carry *Salmonella* between flocks, bypassing the regular cleaning and disinfecting of the pens (*40*, *63*). *Salmonella* contamination can also be brought in on the boots and clothing of farm workers, and one study showed that 20.0% of farm workers' boots were contaminated by *Salmonella* (*40*). Non-chlorinated water supplies as well as feed ingredients are potential vectors for *Salmonella* contamination of the flock (*27*, *11*), with less than one CFU/g of feed able to colonize broilers (*79*). Once *Salmonella* has contaminated the broiler it can survive during the entire rearing process. Colonizing in the mucosal surfaces of the gut, it forms a commensal relationship with the broiler. There are usually no visible signs of *Salmonella* infection in the broiler, making it difficult for farm workers to remove the contaminated birds.

Typically *Salmonella* presents lower cell counts in broilers than *Campylobacter* contamination. However the same risks for cross-contamination still apply. At the end of the

rearing stage, proper feed removal time is again important in preventing cross-contamination of *Salmonella* during transport and evisceration. Cross-contamination can occur during transport, with crate contamination being a risk factor for bird exposure, but can also bring *Salmonella* back to a clean farm environment (*50*). Even when the levels of *Salmonella* on transport crates may be too low to recover, there is still a risk (*68*). *Salmonella* has also been recovered among the cleaned crate racks and can survive in the scalder tanks in processing plants (*50*), both of which increase the potential for cross-contamination at the processing plant. Low *Salmonella* recovery on subsequent control points (such as the inside-outside bird washers) may also be due to the removal of the bacteria from broiler carcasses during earlier control points and not as a result of the current equipment cleaning methods (*50*).

4. Pathogen Control

4.1 In Farm and Transport

Raising *Campylobacter*- and *Salmonella*-free broiler flocks is the ideal prevention method to prevent initial contamination of the birds. Current methods to prevent or lower pathogenic growth in poultry include the use of probiotics, vaccinations, and antibiotics. Early probiotic use in broiler chick diets has shown a reduction in the *Salmonella* levels in the ceca and liver, suggesting competitive exclusion may help lower contamination rates of broilers (*10*). Maternal vaccination against *Salmonella* has also shown a reduction in *Salmonella* levels in the liver (*10*). Oral vaccination against *Campylobacter jejuni* showed large reductions in counts in the vaccinated chick ceca (*60*), however, the use of antibiotics in poultry has reduced greatly, as flouroquinolone resistant strains of *Campylobacter jejuni* emerged following the use of the

antibiotic in poultry (*58*). Likewise, the use of antibiotics in poultry production has led to sulfadiazine, neomycin, tetracycline, and streptomycin resistant strains of *Salmonella* Enteritidis in Spain (*21*).

Proper cleaning and disinfection of the broiler may help prevent subsequent flocks from being exposed to *Campylobacter* strains left by previous flocks (*76*). In addition, having separate boots and clothing stored within each broiler house may help prevent cross-contamination of *Campylobacter* due to human exposure (*76*). Sanitization of the chick eggs may help prevent *Salmonella* contamination from penetrating the egg shell or attaching to the transport crates (*18*). Proper cleaning and disinfection of the breeder houses, hatcheries and transport vehicles can prevent *Campylobacter* and *Salmonella* from being brought to the broiler houses. Proper insect control and rodent control may prevent contamination of the litter, feed and water supplies by pest feces. Chlorination of the water supply may be effective against *Salmonella* and *Campylobacter* contamination; however in low levels it has shown no significant difference when compared to unchlorinated water (*74, 59*).

4.2 In Plant Processing Equipment

The scalder is the first point of reduction in microbial load in a poultry processing plant. The main purpose of the scalder is to loosen the feather follicles of the broilers to allow for easy removal of the feathers further down the processing line. Significant reductions of *Campylobacter* on broiler carcasses have been observed when pre-scald carcasses were compared to post scald carcasses after a hard scald rinse in a three-stage counterflow tank (*16*). Reductions in *Campylobacter* and *Salmonella* are also observed when water samples are

compared after each stage in a three-stage counterflow tank (*22*). When chemicals such as sodium hydroxide or a copper sulfate sanitizer are added to the scald water, further reductions in *Salmonella* may be observed (*41*, *65*). Increasing the pH can reduce the prevalence and count of *Campylobacter* on broiler carcasses (*17*). Increasing the temperature may also reduce bacterial survival in the scald water (*69*); however any reduction in bacterial load may be short lived if intestinal contents are released during evisceration (*80*). In addition, too high of a scalder temperature may result in epidermis damage, exposing underlying tissues and allowing foodborne pathogens to adhere to tissues (*69*).

After evisceration, high pressure carcass washers are used as physical means of fecal and bacterial removal in poultry processing plants. The three most common types of carcass washers in poultry processing plants are brush washers, cabinet washers, and IOBW (*34*). Brush washers work by using high pressure sprays and rubber fingers moving at a high speed to remove any fecal matter and other debris from the outside of the broiler carcass. Cabinet washers are similar to brush washers; however only high pressure spray nozzles are used to remove debris from the outside of the broiler carcass. IOBW use an internal probe to enter the bird carcass, using a high pressure spray to remove debris and fecal matter from the inside of the carcass in addition to the outside sprays. If an antimicrobial is used in these cabinets, it is typically chlorine in a concentration of 30 to 50 ppm (not to exceed 20 ppm). While these carcass washers may remove fecal matter and debris from the carcass, the reduction of food pathogens has varied when tested. One study showed that using IOBW resulted in a 2 log CFU/mL reduction of *Salmonella* counts (*48*) whereas, another study displayed a less than 1 log CFU/mL reduction on *Campylobacter* counts (*12*).

As per USDA standards, broiler carcasses are required to be cooled rapidly after evisceration to prevent bacterial growth. This is usually done using a pre-chiller and an immersion primary chiller in the processing plant. The pre-chiller the removal of any blood or organic matter left on the bird prior to entering the primary chiller with a broiler carcass typically in the pre-chiller water for 30 minutes to 1 hour. Some plants use a direct addition of an antimicrobial to the pre-chiller to reduce bacterial load, however most plants use the primary chiller inflow to add any antimicrobial to the water. After exiting the pre-chiller, broiler carcasses enter the primary chiller tank. The carcass usually spends 1 to 2 hours in the primary chiller, and the carcass temperature is brought down to <40°F within 4 hours of evisceration/slaughter. Antimicrobial interventions are focused on this step of processing in most plants, because of extended contact time with the carcass. Agitation, countercurrent flow, and a higher concentration of the preferred antimicrobial are used to reduce the bacterial load on a broiler carcass as much as possible. Studies have shown a significant reduction in *Campylobacter* when antimicrobials are used during the primary chilling process (*52*). Typically chlorine or peroxyacetic acid are used as the antimicrobial; however the antimicrobial and concentration used can vary between plants.

Alternatively, a plant may use an air-chilling system to reduce broiler carcass temperatures. An increased risk of cross-contamination of *Campylobacter* and *Salmonella* in immersion chiller water has been shown when compared to air chilling (*66*), making air chilling a preferred choice in some countries. Antimicrobials are not usually used during air chilling, making the process less of a focus for bacterial load reduction.

While studies have shown that the primary chilling process significantly reduces the bacterial load, many carcasses still exit the chiller with positive numbers of *Campylobacter* still attached (*52*). In an attempt to further reduce the microbial load and lower the amount of *Salmonella* and *Campylobacter*-positive carcasses, the use of a post-chill dip tank or finishing chiller has recently gained popularity in poultry processing plants throughout the United States. A post-chill dip tank finishing chiller uses a higher concentration of antimicrobials than in the primary chiller (usually 2 to 3 times greater), but reduces the contact time of the broiler carcass to 30 seconds in order to avoid quality defects and high chemical residue. The use of these tanks is relatively new to the industry; however one study showed a reduction of both *Salmonella* and *Campylobacter* on broiler carcasses when several different antimicrobials were used in a post-chill immersion tank (*45*). In addition to a post-chill dip tank or finishing chiller, some poultry processing plants also use either a post-chill spray cabinet or post-chill spray bar as a final intervention/corrective action step; however the effectiveness of these interventions has not been reviewed.

4.3 Antimicrobials

Chlorine. Chlorine has been used for over 50 years in the poultry industry as an antimicrobial, and is still one of the most popular and effective methods of pathogen reduction on poultry carcasses. Low cost and availability make chlorine an easy choice as an antimicrobial in immersion chillers and other large tanks. Sodium hypochlorite, a common source of chlorine, forms hypochlorous acid when dissolved in water. The hypochlorous acid then reacts with the bacterial cell, shutting down vital systems and killing the cell. The USDA has the currently

acceptable levels of total chlorine at 20 ppm in bird washers and 50 ppm incoming potable water to the primary chiller, and 5 ppm in the reuse water, or red water entering the pre-chiller (*8*).

The efficacy of chlorine is dependant on several factors, including the pH of the water, the organic matter in the water, and the contact time with the bacterial cell. An ideal pH for chlorine is 6.0, where chlorine almost completely hydrolyzes into hypochlorous acid. Keeping the pH of the water used in poultry processing near 6.0 can be difficult, and the higher the pH is increased, the less effective chlorine is as an antimicrobial. A study by Blaser et al. (*18*) displayed that at a pH of 6.0, only 0.1 mg/L of free available chlorine was needed to inactivate 99% of three *Campylobacter jejuni* strains after 5 minutes of contact. At a pH of 8.5, only 8% of chlorine hydrolyzes into hypochlorous acid, and so the dosage of chlorine needed to be effective as an antimicrobial is much higher. In primary chilling tanks and any other immersion tank in poultry processing, organic load is one of the biggest factors in reducing the efficacy of chlorine as an antimicrobial. Organic matter and other impurities in the water will react with the chlorine, reducing the amount of chlorine available to form hypochlorous acid. This is known as the chlorine demand of the water, and any chlorine added to the water over the chlorine demand is known as free residual chlorine. The higher amount of free residual chlorine in the water, the more effective the chlorine is as an antimicrobial, however too high of a level of free residual chlorine in the water can produce a hazardous amount of chlorine gas emanating from the immersion tanks. The contact time is also important in determining the effectiveness of chlorine as an antimicrobial. Northcutt and Jones (*47*) found that a free available chlorine concentration of 50 ppm had no effect on *Campylobacter* and *Salmonella*

levels when used in an inside-outside bird washer with a contact time of 5 seconds. Mead et al. (*42*) also observed that 10-20 ppm of chlorine had little to no effect on carcass contamination when used in a post-evisceration spray. When used in immersion chillers, the longer contact time of the chlorine is counteracted by the amount of organic matter in the water, which reduces the free available chlorine and lessens its effectiveness against pathogens such as *Salmonella* (*32*). In addition, Yang et al. (*82*) showed that the antimicrobial effect of chlorine against *Campylobacter* is significantly reduced with increased time of chlorine in the water, and Kameyama et al. (*33*) showed that the levels of chlorine concentration decreased, the longer it remained in the chiller water.

Peracetic Acid (PAA). Peracetic acid is quickly becoming one of the most used antimicrobials in the poultry industry. PAA (or PAHP) is typically introduced into the water as a combination of peracetic acid and hydrogen peroxide creating equilibrium in the water. The mixture works as both an oxidizer and acid, disrupting the permeability of the cell membrane and altering protein synthesis. The current USDA standards for peracetic acid use are 220-ppm peracetic acid, 110-ppm hydrogen peroxide in sprays and chiller water, and 2000-ppm peracetic acid in a post-chill dip (*8*).

The efficacy of peracetic acid has been shown to be strong in several applications. At 85 ppm in chiller water, PAA reduced *Salmonella*-positive carcasses by 92% and *Campylobacter*-carcasses by 43% (*14*). Chantarapanont et al. (*23*) also found that the level of *Campylobacter jejuni* on chicken skin was significantly reduced when submerged in a solution containing 100 ppm of peracetic acid for 15 minutes. *Salmonella*-positive broiler carcasses showed a 2.1 log

CFU/mL reduction and *C. jejuni*-positive carcasses showed a 2.0 log CFU/mL reduction when exposed to 1000 ppm of PAA for 20 seconds in a post-chill immersion tank (*45*). Bauermeister et al. (*14*) also showed that 200-ppm PAA had a 1 log CFU/mL greater reduction of *Salmonella* on broiler carcasses when compared to 30 ppm of chlorine after a 1 hour chiller exposure. There was also little to no quality loss when peracetic acid-treated poultry samples were compared to control samples in a sensory panel (*14*).

Trisodium Phosphate (TSP). TSP is a mixture typically used as a spray in online reprocessing (OLR) or carcass rinse cabinets. When TSP comes in contact with a carcass, the antimicrobial reduces bacterial counts through its high pH of 11.0, which removes some of the surface fat on the carcass (containing bacteria), removing attached surface bacteria, and disrupting the bacterial cell membrane. The USDA has approved the use of a 12% solution of TSP on poultry carcasses; however there are several negative effects to its use. When residual TSP on the poultry carcasses enters the immersion tanks, it raises the pH of the water, reducing the antimicrobial effectiveness of any other chemicals (such as chlorine) used in conjunction. The cleaning of the carcasses to prevent this buildup can be costly for a company (*3*). In addition the phosphate concentration of the waste water of a plant increases, requiring further methods of action to prevent environmental pollution (*51*).

Despite these negative effects, TSP is an effective antimicrobial against many foodborne pathogens. Somers et al. (*72*) showed a reduction of both *Campylobacter* and *Salmonella* biofilms when exposed to 8% TSP for 2 minutes. Hwang (1995) showed a significant reduction in viable *Salmonella* cells on chicken skin washed with a 1% TSP solution for 30 minutes.

Rodriguez de Ledesma et al. (*62*) showed a 93.45% reduction in *Salmonella* Typhimurium on chicken wings when exposed to 10% TSP for 15 seconds. Li et al. (*39*) showed a 3.7 log CFU/mL reduction of *S.* Typhimurium when chicken carcasses were sprayed with 10% TSP for 5 seconds. Whyte et al. (*81*) showed a 1.71 log CFU/g reduction in *Campylobacter* on poultry neck skin when dipped in a 10% TSP solution for 15 seconds.

Cetylpyridinium Chloride (CPC). CPC is a quaternary ammonium compound (typically used in oral antiseptics) recently approved for use to treat the raw surface of poultry carcasses. CPC attacks the foodborne pathogens by increasing the permeability of the cell wall, causing lysis and reduced attachment to the poultry skin. Due to the nature of the compound, the USDA-FSIS has only approved the use of 0.3 g CPC per pound of poultry. If it is used in a dip, the dwell time cannot be longer than 10 seconds and 99% of the solution must be recycled from the carcass, usually through a secondary rinse (*8*). This is to prevent a toxic effect to consumers due to the consumption of CPC residue left on poultry carcasses.

The effectiveness of CPC as an antimicrobial has been demonstrated in both *Campylobacter* and *Salmonella*. Arritt et al. (*9*) displayed a 2.89 log CFU/mL reduction in *C. jejuni* levels on chicken skin when 0.5% CPC was applied to the sample. Li et al. (*39*) demonstrated a 1.6 log CFU/mL reduction of *Salmonella* Typhimurium when 0.1% CPC was applied for 90 seconds to broiler carcasses. Beers et al. (*15*) showed that 0.5% CPC applied to pre-chill broiler carcasses, reduced *Campylobacter* and *Salmonella* incidence from 98% and 34% on untreated carcasses to 8% and 9% on CPC treated carcasses, respectively.

Acidified Sodium Chlorite (ASC). Acidified sodium chlorite is typically used as a disinfectant in the medical field, and was approved by the USDA for use as an antimicrobial in poultry production in 1999. ASC works in a pH range of 2.2-3.0 and oxidizes the bacterial cell wall, attacking disulfide links and the amino acids located in the cell wall. Applied as a spray or dip prior to chiller immersion in poultry production, ASC is approved so that the acid used (typically citric acid) reduces the pH of the solution to a low level (2.0-3.0) and the sodium chlorite concentration is limited at 1200 ppm (*8*). When used in a pre-chiller or primary chiller, the sodium chlorite limit is reduced to 50-150 ppm.

Kemp et al. (*36*) found that when used as a pre-chill spray, fecal contamination was removed and a *Campylobacter* reduction of 1.75 log CFU/mL was observed compared to carcasses that were taken off-line for reprocessing. Kemp et al. (*36*) also found that poultry carcasses treated online with an ASC system reduced the incidence of *Salmonella* from 31.6% to 10.0% when compared to standard offline reprocessing procedures. Oyarzabal et al. (*52*) observed a reduction of *Campylobacter* concentrations to less than 0.2 log CFU/mL on carcasses treated with ASC.

Chlorine Dioxide. Chlorine dioxide (ClO_2) as an antimicrobial has been used as a gas disinfectant for over a century, and in 1999 the FDA approved the compound in an aqueous solution for use in poultry processing water and it is currently approved in immersion tanks with a maximum residual chlorine dioxide level of 3 ppm (*8*). Chlorine dioxide has an oxidizing power 2.5 times stronger than chlorine, is less affected by pH change, and does not react with ammonia. Similar to ASC, it disrupts the permeability of the bacterial cell membrane,

preventing protein synthesis and preventing nutrient transport. The smaller dosage required for effective microbial control makes it a cost-effective option for processing companies, however there may be a slight bleaching effect on poultry skin (*75*). An experiment by Smigic et al. (*70*) showed that 97% of *Campylobacter jejuni* cells were injured when exposed to 20 ppm ClO_2 for 2 minutes.

Irradiation. Irradiation is a physical treatment of the poultry carcass designed to attack the critical bacterial cell components and damage the DNA to a point of cell inactivation and death. Unlike other antimicrobial treatments irradiation can penetrate all areas of the poultry carcass, making it a more thorough intervention. The USDA has currently approved irradiation for use in poultry production at a maximum dose of 3 kGy, however it is not yet widely used in the industry. Patterson (*54*) found that a radiation dose of 2.5 kGy was sufficient in eliminating several *Campylobacter* strains of up to 10.0 log CFU/mL on poultry meat.

5. Summary

Foodborne illness caused by *Salmonella*- and *Campylobacter*- contaminated poultry continues to be an issue in the industry despite stricter regulations and advancement in detection methods. Current on-farm prevention techniques show some effect in the reduction of *Salmonella* and *Campylobacter* in broiler flocks, but due to the large amount of contamination sources and the rapid spread within a flock elimination of the pathogens at the farm level may be impractical. Many current processing plant antimicrobials have shown reductions in different stages of the processing line; however *Salmonella* and *Campylobacter* contamination of the broiler carcass still remains an issue in post-processing and retail. Due to

current consumer perception and up-front expenses, more effective antimicrobial treatments such as gamma irradiation remain scarce in industry usage. Many current antimicrobial treatments, including chlorine and PAA are also banned in several countries, forcing processing plants to use less effective antimicrobials to increase export sales. In Europe, most in-plant chemical treatments of broiler carcasses are banned, leaving it up to the farm level to prevent *Salmonella* and *Campylobacter* contamination from reaching the processing plant. Proper consumer education, the development of cost-effective antimicrobials, and setting global industry practice standards should be the focus to assist in the global reduction of poultry-related foodborne illness.

Chapter 2: Evaluating the Best Management Practices for *Campylobacter* and *Salmonella* Reduction in Southeastern Poultry Processing Plants

2.1 Introduction

Contaminated poultry products are the cause of over 1.5 million cases of foodborne illness annually in the United States, resulting in over $2.5 billion in cost of illness. *Campylobacter* and *Salmonella* are the top two pathogens associated with poultry-related foodborne illness, accounting for 800,000 cases of infection alone (*13*). From 2006 to 2011, the incidence of *Campylobacter* infection had increased 14% and the incidence of *Salmonella* infection had increased 6%, with poultry continuing to be a primary reservoir for the pathogens (*6*). To help reduce the incidence of poultry-related foodborne illness, new performance standards for poultry processing plants were implemented in 2011 by the USDA-FSIS. The new standards included reducing the acceptable number of *Salmonella*-positive samples in a 51-sample set to five and the acceptable number of *Campylobacter*-positive samples in a 51-sample set to eight. Immediate follow-up testing will occur if a plant fails initial testing, and based on the follow-up test results the FSIS may Web-post individual plants that failed testing for public viewing. If current performance trends continue to decline, then further action may be taken in the future (7).

Salmonella and *Campylobacter* form a commensal relationship with the chicken, making contamination difficult to detect at the farm level. In addition, the spread of the pathogens within flocks can be rapid due to the caprophagic nature of the bird, as well as the transfer of the pathogens through outside sources such as communal water and farm employees. Transportation of the live birds to the processing plant is another source of cross-contamination as the transport crates may be poorly sanitized between flocks. Weather conditions and holding

times are also potential causes of cross-contamination of poultry during transport. High temperatures can cause heat stress among, which in addition to longer holding times can lead to increased fecal shedding among flocks. Due to these issues with live birds, poultry processing companies are adjusting their in-plant intervention methods to meet the stricter regulation guidelines.

Currently there are no standard best management practices for pathogen reduction in poultry processing plants. Zero fecal contamination and <40°F carcass temperature exiting the chiller are required critical control points regulated by the USDA. One aspect that varies greatly between plants is the antimicrobials treatments used to reduce *Salmonella* and *Campylobacter* load on poultry carcasses.

While the antimicrobial treatments used in each plant may vary greatly, scalder tanks, inside-outside bird washers (IOBW), a pre-chiller, and a primary immersion chiller are usually the standard intervention points. However the number of scalders, number of pre-chillers, and whether or not a post-chill intervention (such as a finishing chiller or dip tank) is used in the production line can vary between plants. Immersion time of the poultry carcass in the pre-chiller and primary chiller is also non-standard between plants.

With all of the intervention options available to processing plants, the focus of this study was to determine which management practices were best for reducing the number of *Campylobacter* and *Salmonella*-positive broiler carcasses exiting the production line, as well as reducing the microbial load of the carcass. In determining these best management practices poultry processing plants can adjust their production lines to meet and exceed USDA standards.

The resulting practices would reduce the costs of production as well as ensure the consumer that safe poultry products are exiting the processing plant.

2. Materials and Methods

Surveys. Surveys were sent out to the QA managers of eight processing plants in the Southeastern U.S. to assess the plants current production size, cleaning methods, operation setup, antimicrobials used, and the foodborne pathogens regularly tested on-site. Based on the results of the surveys, six out of the eight processing plants were identified for sampling and the initial sampling procedures were planned to fit each processing plant. The sampling focused on the intervention points along the processing line as well as several points of possible cross-contamination.

Initial Plant Sampling. After the surveys were completed, sampling kits for six of the processing plants were prepared. Each plant was sampled twice during the week (Monday and Thursday) during the winter months (November-February) for microbial analysis. Carcass rinse samples were taken before the inside-outside bird washers in the plant, before the broiler carcasses entered the pre-chiller, between the pre-chiller treatment and primary chiller treatment, after the primary chiller treatment, and after the post-chill antimicrobial treatment (if applicable). In addition, water samples were taken from each scalder, the pre-chiller(s), the primary chiller, and the finishing chiller or dip tank (if applicable) using a collector scoop. Finally, a manure grab was taken from the end of the shackling belt to assess incoming microbial load. A total of 25 bird rinses, made up of 400mL of buffered peptone rinse (Neogen®, Lansing, MI) were prepared and sterilized for transport. Two mL syringes of sodium thiosulfate

were also prepared for each water sample to neutralize any residual antimicrobial. Five broiler carcasses were randomly taken off the production line at each sampling location and combined in a sterile rinse bag with a 400mL buffered peptone rinse. The carcasses were then subjected to a 1 minute carcass rinse, after which they were placed back onto the production line. The rinse was then added to a sterile stomacher bag and sealed for transport. Water samples were collected from all tanks within the processing plants, added to a sterile stomacher bag, and combined (if necessary) with 2mL of sodium thiosulfate to neutralize any residual effect of an antimicrobial. All stomacher bags were packed in ice and transported back to the laboratory at Auburn University for analysis.

Laboratory Preparation and Testing.

Day 1: Once samples were received at the laboratory, the manure sample was diluted using a 1 to 3 dilution of buffered peptone water (BPW) (Neogen®, Lansing, MI) in a stomacher bag, and stomached for 1 minute. Thirty mL of each sample was combined with 30 mL of sterile Bolton broth (Neogen®, Lansing, MI) with 2X strength Bolton broth selective supplement (EMD Chemicals Inc., Gibbstown, NJ) (made up of 10mg vancomycin, 10mg cefoperazone, 10mg trimethoprim, and 5mg amphotericin B) in a stomacher bag and sealed in a microaerophilic environment for 48 hours at 42°C for *Campylobacter* enrichment. Thirty mL buffered peptone was added to 30mL of the water samples and all buffered peptone samples were incubated for 24 hours at 37°C for *Salmonella* enrichment. One mL of each sample was also combined with 9mL of 0.1% peptone (Neogen®, Lansing, MI), for 1 to 10 dilutions, up to a 10^{-2} dilution (manure grab samples were increased to a 10^{-6} dilution). 100-µL of each dilution was plated onto Xylose Lysine Tergitol-4 (XLT-4) (Neogen®, Lansing, MI) agar (4.6mL/L XLT-4 supplement added

(Neogen®, Lansing, MI)) for enumeration of *Salmonella*. XLT-4 plates were incubated for 24 hours at 37°C. Campy-Cefex agar (Neogen®, Lansing, MI) plates (50mL/L laked horse blood (Quad Five, Ryegate, MT) and 0.033g/L Cefoperazone (Toronto Research Chemicals Inc., North York, ON Canada)added) were used for *Campylobacter* enumeration, 100-µL of each sample added to the 10^{-2} and 10^{-1} dilution plates, and 250-µL added to four Campy-Cefex plates (for a total of 1-mL sampled) in compliance with USDA standards. Campy-Cefex plates were incubated in a microaerophilic environment for 48 hours at 42°C. Detection limits for *Campylobacter* enumeration occurred at 1 CFU/mL and detection limits for *Salmonella* enumeration occurred at 1 CFU/200µL.

Day 2: After 24 hours XLT-4 plates were counted and recorded in CFU/mL. 100-µL of each *Salmonella* enrichment buffered peptone samples were transferred into 10mL of Rappaport Vassiliadis (RV) (Neogen®, Lansing, MI) broth and incubated at 37°C for an additional 24 hours.

Day 3: After 48 hours Campy-Cefex plates were counted and recorded in CFU/mL. Each *Campylobacter* enrichment sample in Bolton broth was streaked onto a Campy-Cefex plate and incubated at 42°C for 48 hours to test for *Campylobacter* prevalence. Each *Salmonella* enrichment sample in RV broth was then streaked onto a XLT-4 plate and incubated at 37°C for 24 hours to test for *Salmonella* prevalence.

Day 4: After 72 hours XLT-4 streak plates were analyzed for *Salmonella* prevalence and any positive samples were recorded.

Day 5: After 96 hours Campy-Cefex streak plates were analyzed for *Campylobacter* prevalence and any positive samples were recorded.

Initial Sampling Analysis and Secondary Sampling. All enumeration samples and all positive *Campylobacter* and *Salmonella* samples in each plant were recorded and intervention methods were compared. Based on the results, changes to one or more intervention methods were recommended to each processing plant, following which a secondary sampling set was performed at each processing plant during the summer months (May-August), and adjustments were made to the amount of samples taken based on the changes made within each plant. After the second set of samples were taken, analysis within each sampling set and between the first and second sampling set to determine microbial loads, the effect of the changes, and the effect of seasonal weather on microbial loads were performed.

3. Results

Note: In all plants, *Salmonella* survival was below detectable levels in all carcass rinse samples, water samples and the manure grab samples (Detection limit = 1 CFU/200µL).

Plant A: Analysis of microbial load and buildup. The production line of Plant A included 3 scalder tanks, an inside-outside bird washer (IOBW), two pre-chillers (a 20 minute rocker pre-chiller and a 30 minute screw pre-chiller), a 90 minute primary chiller, and a post-chill rinse bar system. Chlorine was used as the antimicrobial in the IOBW (50 ppm), two pre-chillers (50 ppm), primary chiller (50 ppm), and post-chill rinse bar system (30 ppm). Initial carcass samples from Plant A were taken before entering the IOBW, after exiting the IOBW, after exiting the 2nd pre-chiller, after exiting the primary chiller, and after exiting the post-chill rinse bars. Water samples were taken from the three scalder tanks, the two pre-chillers, and the primary chiller. A manure grab was taken from the end of the shackling belt.

When the 1st Monday sampling period was compared to the 1st Thursday sampling period (Fig. 1.1), the 1st Thursday bird samples displayed significantly ($p<0.05$) higher *Campylobacter* counts after the IOBW, increasing from 0.79 Log CFU/mL to 1.71 Log CFU/mL. When the 2nd Monday sampling period was compared to the 2nd Thursday sampling period (Fig. 1.1), the 2nd Thursday bird samples displayed significantly ($p<0.05$) higher *Campylobacter* counts post pick and after exiting the pre-chiller, increasing from 1.59 Log CFU/mL to 2.90 Log CFU/mL and 1.18 Log CFU/mL to 1.88 Log CFU/mL respectively. The 2nd Thursday pre-chiller water sample (Fig. 1.2) also displayed a higher survival rate of *Campylobacter*, increasing from 1.28 Log CFU/mL to 2.55 Log CFU/mL.

Effectiveness of changes made within the plant. Changes that were made at the plant included removing one of the pre-chillers, changing from a post-chill rinse bar system to a post-chill dip tank, and switching from chlorine to peracetic acid (PAA) as the antimicrobial used in the primary chiller (10 ppm) and post-chill dip tank (100 ppm). The chemicals used in the scalder were also removed. As a result of the changes, the 2nd pre-chiller water sample was removed and a post-chill dip tank water sample was added.

When the 1st Monday sampling period was compared to the 2nd Monday sampling period (Fig 1.1), the 2nd Monday bird samples displayed significantly ($p<0.05$) higher *Campylobacter* counts after exiting the IOBW and after exiting the pre-chiller, increasing from 0.79 Log CFU/mL to 1.40 Log CFU/mL and 0 CFU/mL to 1.18 Log CFU/mL respectively. When the 1st Thursday sampling period was compared to the 2nd Thursday sampling period (Fig1.1), the 2nd Thursday bird samples displayed significantly ($p<0.05$) higher *Campylobacter* counts after exiting the pre-chiller, increasing from 0.19 Log CFU/mL to 1.88 Log CFU/mL.

***Salmonella* Enrichments.** In the 1st sampling set, *Salmonella* was detected in two scalder water samples on Monday and Thursday as well as the Thursday pre-chiller water sample and one post-chill bird sample (Table 1.1).

In the 2nd sampling set, *Salmonella* was detected in the manure grab sample on Monday and Thursday as well as the 1st scalder tank on Monday.

Plant B: Analysis of microbial load and buildup. The production line of Plant B included 3 scalder tanks, an inside-outside bird washer (IOBW), one 20 minute pre-chiller, and a 120 minute primary chiller. No post chill intervention was used, with a spray bar only used for corrective actions. Chlorine was used as the antimicrobial in the IOBW (50 ppm), and peracetic acid (PAA) was used as the antimicrobial in the pre-chiller (residual concentration from primary chiller overflow) and primary chiller (25 ppm). Initial carcass samples were taken before entering the IOBW, after exiting the IOBW, after exiting the pre-chiller, and after exiting the primary chiller. Water samples were taken from each scalder tank, the pre-chiller, and the primary chiller. A manure grab was taken from the end of the shackling belt.

When the 1st Monday sampling period was compared to the 1st Thursday sampling period (Fig. 2.1), the 1st Thursday bird samples displayed significantly ($p<0.05$) higher *Campylobacter* counts after exiting the IOBW and after exiting the pre-chiller, increasing from 0.06 Log CFU/mL to 0.65 Log CFU/mL and 0.16 Log CFU/mL to 0.79 Log CFU/mL respectively. The 1st Thursday pre-chiller water sample (Fig. 2.2) also displayed higher *Campylobacter* survival, increasing from 0.48 Log CFU/mL to 1.15 Log CFU/mL. When the 2nd Monday sampling period was compared to the 2nd Thursday sampling period (Fig. 2.1) bird samples displayed no significant ($p\geq0.05$) difference in *Campylobacter* load.

Effectiveness of changes made within the plant. Changes made included adding a finishing chiller after the primary chiller, increasing the scald time of the birds, and switching from PAA to bromine as the antimicrobial in the primary chiller (10 ppm) and finishing chiller (140 ppm). As a result of the changes, five bird samples were added after exiting the finishing chiller, and a water sample was added from the finishing chiller.

When the 1st Monday sampling period was compared to the 2nd Monday sampling period (Fig. 2.1), the 2nd Monday bird samples displayed significantly ($p<0.05$) higher *Campylobacter* counts post-pick and after exiting the IOBW, increasing from 0.53 Log CFU/mL to 1.78 Log CFU/mL and 0.06 Log CFU/mL to 1.55 Log CFU/mL respectively. The 2nd Monday manure grab sample (Fig. 2.2) also displayed a higher initial *Campylobacter* load, increasing from 4.92 Log CFU/mL to 6.29 Log CFU/mL. When the 1st Thursday sampling period was compared to the 2nd Thursday sampling period (Fig. 2.1) bird samples displayed no significant ($p\geq0.05$) difference in *Campylobacter* counts.

***Salmonella* Enrichments.** In the 1st sampling set, *Salmonella* was detected in the manure grab, 1st and 3rd scalder tank water samples on both Monday and Thursday (Table 2.1). On Monday the 2nd scalder tank water sample, pre-chiller water sample, three post-pick bird samples, three pre-chill bird samples, and four pre-post bird samples also tested positive for *Salmonella*. On Thursday the primary chiller water sample tested positive for *Salmonella.*

In the 2nd sampling set, *Salmonella* was detected in the 2nd scalder tank water sample on both days. On Monday the 3rd scalder tank water sample, pre-chiller water sample, two post pick bird samples, and one pre-chill bird sample tested positive for *Salmonella*. On Thursday the

1st scalder tank water sample, four pre-chill bird samples, and one pre-post bird sample tested positive for *Salmonella*.

Plant C: Analysis of microbial load and buildup. The production line of Plant C included two scalder tanks, an inside-outside bird washer (IOBW), one 15 minute pre-chiller, a 60 minute primary chiller, and a post-chill dip tank. Chlorine was used as the antimicrobial in the IOBW (40 ppm), pre-chiller (residual concentration from primary chiller overflow), primary chiller (35 ppm), and post-chill dip tank (60 ppm). Carcass samples were taken before entering the IOBW, after exiting the IOBW, after exiting the pre-chiller, after exiting the primary chiller, and after exiting the post-chill dip tank. Water samples were collected from the two scalder tanks, the pre-chiller, the primary chiller, and the post-chill dip tank. A manure grab was taken from the end of the shackling belt.

When the 1st Monday sampling period was compared to the 1st Thursday sampling period (Fig. 3.1), the 1st Thursday bird samples displayed significantly ($p<0.05$) lower *Campylobacter* counts post-pick, after exiting the IOBW, after exiting the pre-chiller, after exiting the primary chiller, and after exiting the dip tank, decreasing from 2.77 Log CFU/mL to 0.96 Log CFU/mL, 2.37 Log CFU/mL to 0.76 Log CFU/mL, 1.97 Log CFU/mL to 0.59 Log CFU/mL, 0.62 Log CFU/mL to 0.12 Log CFU/mL, and 0.59 Log CFU/mL to below detection limit respectively. The 1st Thursday manure sample (Fig. 3.2) also displayed a lower initial *Campylobacter* load, decreasing from 7.12 Log CFU/mL to 4.67 Log CFU/mL and all water samples (Fig. 3.2) displayed a lower survival rate of *Campylobacter*. When the 2nd Monday sampling period was compared to the 2nd Thursday sampling period (Fig. 3.1), the 2nd Thursday bird samples displayed significantly ($p<0.05$) higher *Campylobacter* counts after exiting the

IOBW, after exiting the pre-chiller, after exiting the primary chiller, and after exiting the dip tank, increasing from 0.59 Log CFU/mL to 1.62 Log CFU/mL, 0.63 Log CFU/mL to 1.59 Log CFU/mL, undetectable levels to 0.91 Log CFU/mL, and 0.16 Log CFU/mL to 0.75 Log CFU/mL respectively.

Effectiveness of changes made within the plant. A rehang area and Cecure® CPC spray cabinet (.15% CPC) were both added after the post-chill dip tank. As a result of the change, 3 additional bird samples were taken after exiting the CPC spray cabinet.

When the 1st Monday sampling period was compared to the 2nd Monday sampling period (Fig. 3.1), the 2nd Monday bird samples displayed significantly ($p<0.05$) lower *Campylobacter* counts post-pick, after exiting the IOBW, after exiting the pre-chiller, and after exiting the primary chiller, decreasing from 2.77 Log CFU/mL to 1.70 Log CFU/mL, 2.37 Log CFU/mL to 0.59 Log CFU/mL, 1.97 Log CFU/mL to 0.63 Log CFU/mL, and 0.62 Log CFU/mL to 0 CFU/mL respectively. The 2nd Monday manure sample (Fig. 3.2) also displayed a lower initial *Campylobacter* load, decreasing from 7.12 Log CFU/mL to 5.47 Log CFU/mL. When the 1st Thursday sampling period was compared to the 2nd Thursday sampling period (Fig. 3.1), the 2nd Thursday bird samples displayed significantly ($p<0.05$) higher *Campylobacter* counts post-pick, after exiting the IOBW, after exiting the pre-chiller, after exiting the primary chiller, and after exiting dip tank, increasing from 0.96 Log CFU/mL to 1.45 Log CFU/mL, 0.76 Log CFU/mL to 1.62 Log CFU/mL 0.59 Log CFU/mL to 1.59 Log CFU/mL, 0.12 Log CFU/mL to 0.91 Log CFU/mL, and undetectable levels to 0.75 Log CFU/mL respectively. The 2nd Thursday manure sample (Fig. 3.2) also displayed a higher initial *Campylobacter* load, increasing from 4.67 Log CFU/mL to 5.36 Log CFU/mL.

***Salmonella* Enrichments.** In the 1st sampling set, no *Salmonella* was detected on any of the bird samples, water samples, or manure grab (Table 3.1). In the 2nd sampling set, on Thursday the dip tank water sample, one pre-chill bird sample, and one pre-post bird sample tested positive for *Salmonella*.

Plant D: Analysis of microbial load and buildup. The production line of Plant D included three scalder tanks, an inside-outside bird washer (IOBW), a 20 minute pre-chiller, a 120 minute primary chiller, and no post chill intervention. Chlorine was used as the antimicrobial in the IOBW (20 ppm), the pre-chiller (residual concentration from primary chiller overflow), and the primary chiller (50 ppm). Carcass samples were taken before entering the IOBW, after exiting the IOBW, after exiting the pre-chiller, and after exiting the primary chiller. Water samples were taken from the three scalder tanks, the pre-chiller, and the primary chiller. A manure grab was taken at the end of the shackling belt.

When the 1st Monday sampling period was compared to the 1st Thursday sampling period (Fig. 4.1), the 1st Thursday bird samples displayed significantly ($p<0.05$) lower *Campylobacter* counts post-pick, after exiting the IOBW, after exiting the pre-chiller, and after exiting the primary chiller, decreasing from 2.80 Log CFU/mL to 1.81 Log CFU/mL, 3.12 Log CFU/mL to 1.93 Log CFU/mL, 3.04 Log CFU/mL to 1.55 Log CFU/mL, and 1.04 Log CFU/mL to 0.16 Log CFU/mL respectively. The 1st Thursday manure sample (Fig. 4.2) also displayed a higher initial *Campylobacter* load, increasing from 5.60 Log CFU/mL to 6.11 Log CFU/mL. When the 2nd Monday sampling period was compared to the 2nd Thursday sampling period (Fig. 4.1), the 2nd Thursday bird samples displayed significantly ($p<0.05$) lower *Campylobacter* counts post-pick and after exiting the IOBW, decreasing from 1.45 Log CFU/mL to 0.83 Log CFU/mL and 0.96 Log

CFU/mL to 0.06 Log CFU/mL respectively. The 2nd Thursday manure sample also displayed a lower initial *Campylobacter* load, decreasing from 4.59 Log CFU/mL to 4.02 Log CFU/mL.

Effectiveness of changes made within the plant. No changes were made between sampling periods.

When the 1st Monday sampling period is compared to the 2nd Monday sampling period (Fig. 4.1), the 2nd Monday bird samples displayed significantly ($p<0.05$) lower *Campylobacter* counts post-pick, after exiting the IOBW, after exiting the pre-chiller, and after exiting the primary chiller, decreasing from 2.8 Log CFU/mL to 1.45 Log CFU/mL, 3.12 Log CFU/mL to 0.96 Log CFU/mL, 3.04 Log CFU/mL to 0.06 Log CFU/mL, and 1.04 Log CFU/mL to 0.16 Log CFU/mL respectively. The 2nd Monday manure sample (Fig. 4.2) also displayed a lower initial *Campylobacter* load, decreasing from 5.60 Log CFU/mL to 4.59 Log CFU/mL. When the 1st Thursday sampling period was compared to the 2nd Thursday sampling period (Fig. 4.1), the 2nd Thursday bird samples displayed significantly ($p<0.05$) lower *Campylobacter* counts post-pick, after exiting the IOBW, and after exiting the pre-chiller, decreasing from 1.81 Log CFU/mL to 0.83 Log CFU/mL, 1.93 Log CFU/mL to 0.06 Log CFU/mL, and 1.55 Log CFU/mL to 0.06 Log CFU/mL respectively. The 2nd Thursday manure sample (Fig. 4.2) also displayed a lower initial *Campylobacter* load, decreasing from 6.11 Log CFU/mL to 4.02 Log CFU/mL.

***Salmonella* Enrichments.** In the 1st sampling set, on Thursday the manure grab tested positive for *Salmonella* (Table 4.1). In the 2nd sampling set, on Thursday one post-pick bird sample tested positive for *Salmonella*.

Plant E: Analysis of microbial load and buildup. The production line of Plant E included 3 scalder tanks, an inside-outside bird washer (IOBW), one 20 minute pre-chiller, one 120

minute primary chiller, and two post-chill spray bars. Chlorine was used as the antimicrobial in the IOBW (35 ppm), the pre-chiller (residual concentration from primary chiller overflow), the primary chiller (50 ppm), and the post-chill spray bars (50 ppm). Carcass samples were taken before entering the IOBW, after exiting the IOBW, after exiting the pre-chiller, after exiting the primary chiller, and after the post chill spray bars. Water samples were taken from the three scalder tanks, the pre-chiller, and the primary chiller. A manure grab was taken from the end of the shackling belt.

When the 1st Monday sampling period was compared to the 1st Thursday sampling period (Fig. 5.1), the 1st Thursday bird samples displayed significantly ($p<0.05$) lower *Campylobacter* counts post-pick, after exiting the IOBW, and after exiting the pre-chiller, decreasing from 1.94 Log CFU/mL to 0.16 Log CFU/mL, 1.1 Log CFU/mL to 0 CFU/mL, and 0.43 Log CFU/mL to 0 CFU/mL respectively. The 1st Thursday manure sample (Fig. 5.2) also displayed a lower initial *Campylobacter* load, decreasing from 6.91 Log CFU/mL to 3.48 Log CFU/mL. When the 2nd Monday sampling period was compared to the 2nd Thursday sampling period (Fig. 5.1), the 2nd Thursday bird samples displayed significantly ($p<0.05$) lower *Campylobacter* counts post-pick, decreasing from 2.43 Log CFU/mL to 1.74 Log CFU/mL.

Effectiveness of changes made within the plant. Changes made include the removal of the post chill spray bars, the addition of citric acid to the third scalder tank, the addition of citric acid to the IOBW spray, and the switch from chlorine to PAA + caustic NaOH as the antimicrobial in the primary chiller (55 ppm). As a result of the changes, the five carcass samples after the spray bars were removed.

When the 1st Monday sampling period was compared to the 2nd Monday sampling period (Fig. 5.1), the 2nd Monday bird samples displayed significantly ($p<0.05$) higher *Campylobacter* counts post-pick, increasing from 1.94 Log CFU/mL to 2.43 Log CFU/mL. When the 1st Thursday sampling period was compared to the 2nd Thursday sampling period (Fig. 5.1), the 2nd Thursday bird samples displayed significantly ($p<0.05$) higher *Campylobacter* counts post-pick, after exiting the IOBW, and after exiting the pre-chiller, increasing from 0.16 Log CFU/mL to 1.74 Log CFU/mL, 0 CFU/mL to 1.32 Log CFU/mL, and undetectable levels to 0.36 Log CFU/mL respectively. The 2nd Thursday manure sample (Fig. 5.2) also displayed a higher *Campylobacter* load, increasing from 3.48 Log CFU/mL to 7.30 Log CFU/mL.

***Salmonella* Enrichments.** In the 1st sampling set, on Monday the manure grab and one post-pick bird sample tested positive for *Salmonella* (Table 5.1). On Thursday the 1st scalder water sample, two pre-chill bird samples, and one pre-post bird sample tested positive for *Salmonella*. In the 2nd sampling set, on both Monday and Thursday the manure grab and 1st scalder water sample tested positive for *Salmonella*. On Monday the 2nd scalder water sample and two post-pick bird samples tested positive for *Salmonella*.

Plant F: Analysis of microbial load and buildup. The production line of plant F included three scalder tanks, an inside-outside bird washer (IOBW), a 20 minute pre-chiller, a 120 minute main-chiller, and a post-chill dip tank. Chlorine was used as the antimicrobial in the IOBW (50 ppm) and PAA was used as the antimicrobial in the main-chiller (8 ppm) and post-chill dip tank (80 ppm). Carcass samples were taken before entering the IOBW, after exiting the IOBW, after exiting the pre-chiller, after exiting the primary chiller, and after exiting the post-chill dip tank.

Water samples were taken from the three scalder tanks, the pre-chiller, the primary chiller, and the post-chill dip tank. A manure grab was taken from the end of the shackling belt.

When the 1st Monday sampling period was compared with the 1st Thursday sampling period (Fig. 6.1), the 1st Thursday bird samples displayed significantly ($p<0.05$) lower *Campylobacter* counts after exiting the IOBW, decreasing from 2.00 Log CFU/mL to 0.65 Log CFU/mL. When the 2nd Monday sampling period was compared to the 2nd Thursday sampling period (Fig. 6.1), the 2nd Thursday bird samples displayed significantly ($p<0.05$) lower *Campylobacter* counts post-pick, decreasing from 2.42 Log CFU/mL to 1.78 Log CFU/mL. The 2nd Thursday manure sample (Fig. 6.2) also displayed a lower initial *Campylobacter* load, decreasing from 5.72 Log CFU/mL to 5.33 Log CFU/mL.

Effectiveness of changes made within the plant. A new scalding tank was put in between sampling periods, no other changes were made.

When the 1st Monday sampling period (Fig. 6.1) was compared to the 2nd Monday sampling period, bird samples displayed no significant ($p\geq0.05$) difference in *Campylobacter* counts. When the 1st Thursday sampling period was compared to the 2nd Thursday sampling period (Fig. 6.1), the 2nd Thursday bird samples displayed significantly ($p<0.05$) higher *Campylobacter* counts after exiting the IOBW, increasing from 0.65 Log CFU/mL to 1.58 Log CFU/mL.

***Salmonella* Enrichments.** In the 1st sampling set, on Monday the manure grab tested positive for *Salmonella* (Table 6.1). On Thursday one pre-chill bird sample tested positive for *Salmonella*. In the 2nd sampling set, on both Monday and Thursday the manure grab and 1st

scalder water samples tested positive for *Salmonella*. On Thursday the 2nd scalder water sample tested positive for *Salmonella.*

4. Conclusions

Plant A. The reduction of *Campylobacter* during all sampling periods can be seen at each intervention point along the production line (Fig. 1.1-1.2). When comparing the Monday and Thursday bird samples for the 1st sampling period, increased *Campylobacter* counts on Thursday at the IOBW suggest microbial build up at that point in the plant possibly due to the equipment not being cleaned thoroughly between production runs. Comparing the Monday and Thursday bird samples for the 2nd sampling period, we observed the same issue on Thursday at post pick and after the pre-chiller, suggesting improper cleaning. The incoming *Campylobacter* load on Thursday was lower than the incoming *Campylobacter* load on Monday, and therefore is not a cause of the increased counts on the Thursday samples. The increase in survival of *Campylobacter* in the pre-chiller water also may be due to residual *Campylobacter* between production runs, and may potentially cross-contaminate the birds. There was only one more positive sample (Table 1.2) post chill, and there was no significant difference in *Campylobacter* levels between the 2nd Monday and Thursday samples after exiting the primary chiller.

Comparing the bird samples of two Monday sampling periods and two Thursday sampling periods, the significantly higher *Campylobacter* counts during the 2nd sampling period after exiting the pre-chiller is most likely due to the removal of one of the pre-chillers between sampling periods, resulting in reduced immersion and antimicrobial exposure time before the

birds were sampled after exiting the pre-chiller. There was no significant ($p \geq 0.05$) difference in *Campylobacter* counts after exiting the primary chiller, however one *Salmonella*-positive bird sample was found after exiting the primary chiller and detectable levels of *Campylobacter* were found in the primary chiller water sample when the plant was using chlorine, whereas no *Salmonella*-positive samples nor detectable *Campylobacter* levels were found after the plant switched to PAA in the primary chiller (Tables 1.1-1.2), suggesting stronger antimicrobial effects of PAA. No *Campylobacter*-positive or *Salmonella*-positive samples were found after the chlorine drip during the 1st sampling period and after the PAA dip tank of the 2nd sampling period, and so both methods may be effective at reducing these pathogens to below-detectable levels. The removal of the scalder chemicals did not show a difference in *Campylobacter* levels in scalder samples, and so the chemicals may have been ineffective to use.

Plant B. The reduction of *Campylobacter* during all sampling periods can be seen at each intervention point along the production line (Fig. 2.1-2.2). Comparing the Monday and Thursday bird samples of the 1st sampling period, higher *Campylobacter* counts on Thursday exiting the IOBW and pre-chiller suggest potential microbial buildup in these areas due to improper cleaning between production runs. Potentially due to the higher microbial load on the Thursday bird samples, countable levels of *Campylobacter* were found on bird samples after exiting the primary chiller and *Salmonella*-positive bird samples were also found after exiting the pre-chiller (Table 2.1). The improper cleaning also may have led to the increase in *Campylobacter* survival in the pre-chiller water sample on Thursday. Comparing the Monday and Thursday bird samples of the 2nd sampling period we see no significant ($p \geq 0.05$) difference in the bird samples between the two days, suggesting that the equipment is being cleaned properly between

production runs. However there is a slight increase in the number of *Salmonella*-positive and *Campylobacter*-positive samples on Thursday's sampling (Tables 2.1-2.2).

Comparing the bird samples of the two Monday sampling periods and the two Thursday sampling periods higher *Campylobacter* counts during the 2nd Monday sampling period post-pick and after exiting the IOBW suggest improper cleaning or cross-contamination, however higher initial *Campylobacter* load counts are also detected from the 2nd Monday manure sample, and so the difference may be due to an increase of *Campylobacter* before reaching the processing plant. There were countable levels of *Campylobacter* in the 2nd Monday sample after exiting the primary chiller as well as in the primary chiller water sample, suggesting that bromine may not be as effective as an antimicrobial as PAA for use in the primary chiller. The number of *Campylobacter*-positive bird samples increased for both the 2nd Monday and 2nd Thursday sampling periods (Table 2.2), further suggesting that bromine may not be as effective as an antimicrobial as PAA. The addition of the dip tank did not strongly affect the *Campylobacter* levels on bird samples during the 2nd Monday sample, however these levels were already very low, and the dip tank was added after the 1st sampling period, so a true analysis of its effectiveness could not be made at this plant.

Plant C. The reduction of *Campylobacter* during all sampling periods can be seen at each intervention point along the production line (Fig 3.1-3.2). Comparing the Monday and Thursday bird samples of the 1st sampling period, significant ($p<0.05$) decreases after each sampling point in the production line as well as in initial *Campylobacter* load may be due to the lower initial load. It also suggests that no microbial buildup occurred to add to the initial *Campylobacter* counts during Thursday's sampling. When the 2nd Monday and 2nd Thursday sampling periods

were compared significantly ($p<0.05$) higher *Campylobacter* counts are observed in Thursday's bird samples along the production line as well as more *Campylobacter*-positive and *Salmonella*-positive bird samples (Tables 3.1-3.2). Since there was no increased initial *Campylobacter* load in the Thursday manure sample, microbial buildup may be the cause early in the production line due to improper cleaning, adding to the initial *Campylobacter* load.

Comparing the bird samples of the two Monday sampling periods and the two Thursday sampling periods significantly ($p<0.05$) lower *Campylobacter* counts on the 2nd Monday sampling period post-pick, after exiting the IOBW, after exiting the pre-chiller, and after exiting the primary chiller may all be due to the lower initial *Campylobacter* load from the 2nd Monday bird samples rather than any changes made. Significantly ($p<0.05$) higher *Campylobacter* counts post-pick, after exiting the IOBW, after exiting the pre-chiller, and after exiting the primary chiller on the 2nd Thursday bird samples once again may be due to the higher initial *Campylobacter* load. The addition of the CPC cabinet resulted in the undetectable levels of *Campylobacter* on bird samples, and eliminated any residual *Campylobacter* surviving the dip-tank treatment, suggesting the use of CPC post chill as a strong antimicrobial. The number of *Salmonella*-positive and *Campylobacter*-positive samples did not change much between the 1st sampling set and 2nd sampling set (Tables 3.1-3.2), but this is expected as there were not any changes along the production line before the CPC cabinet.

Plant D. The reduction of *Campylobacter* during all sampling periods can be seen at each intervention point along the production line (Fig 4.1-4.2). Comparing the Monday and Thursday bird samples of the 1st sampling period significantly ($p<0.05$) lower *Campylobacter* counts on the Thursday bird samples, as well as a higher initial *Campylobacter* load on the Thursday

manure sample, suggests that there is no microbial buildup during the week between production runs. The number of *Campylobacter*-positive and *Salmonella*-postive samples also does not increase during Thursday's sampling period (Tables 4.1-4.2). When the 2nd Monday and Thursday sampling periods are compared significantly ($p<0.05$) lower *Campylobacter* counts on the Thursday bird samples post-pick and after exiting the IOBW once again suggest no microbial buildup between production runs. There is a lower initial *Campylobacter* load on the Thursday manure sample, but it does not appear to affect any potential microbial buildup further down the production line. There are more *Campylobacter*-positive samples on the Thursday sampling period than on Monday (Table 4.2), but negative water samples from the pre-chiller and primary chiller suggest that if there is cross-contamination, it either occurs early along the production line or at the farm level.

Comparing the bird samples of the two Monday sampling periods and the two Thursday sampling periods show significantly ($p<0.05$) lower *Campylobacter* counts along the production line on the bird samples during the 2nd Monday sampling period, however the lower initial *Campylobacter* load observed from the manure may be the cause rather than any changes within the processing plant. This is again observed in the 2nd Thursday bird samples, where *Campylobacter* counts are lower along the production line as well as in the initial *Campylobacter* load taken from the manure sample. The number of *Salmonella*-positive and *Campylobacter*-positive samples does not change much between the two sample sets (Tables 4.1-4.2), with *Campylobacter*-positive samples only decreasing slightly during the 2nd Monday sampling period. Since this plant did not make any changes between the 1st and 2nd sampling

sets, it is expected that initial microbial load will be the cause of any differences in *Campylobacter* counts along the production line.

Plant E. The reduction of *Campylobacter* during all sampling periods can be seen at each intervention point along the production line (Fig 5.1-5.2). Comparing the Monday and Thursday bird samples of the 1st sampling period significantly ($p<0.05$) lower *Campylobacter* counts post-pick, after exiting the IOBW, and after exiting the pre-chiller as well as the negative water samples indicate no microbial buildup between production runs. The lower number of *Campylobacter*-positive samples (Table 5.2) in Thursday's sampling period also suggests no microbial buildup. However the lower initial *Campylobacter* load may be the reason the *Campylobacter* levels along the production line are not even between the Monday and Thursday samples. When comparing the Monday and Thursday bird samples of the 2nd sampling period, the significantly ($p<0.05$) lower *Campylobacter* counts post-pick on the Thursday bird samples, yet higher levels after exiting the IOBW suggest that there may be cross-contamination or improper cleaning between the two sampling points. There are also a higher number of *Campylobacter*-positive bird samples after exiting the pre-chiller and after exiting the primary chiller, further supporting a point of cross-contamination earlier along the production line.

Comparing the bird samples of the two Monday sampling periods and the two Thursday sampling periods showed significantly ($p<0.05$) higher *Campylobacter* counts on the bird samples of the 2nd Monday sampling period. This in addition to the lower initial *Campylobacter* load from the manure sample indicates that the addition of citric acid to the scalder tank may be ineffective at reducing microbial load before the birds reach the post-pick production line.

Higher *Campylobacter* levels were also observed post-pick when the 2nd Thursday was compared to the 1st Thursday sampling period, and while this may be due in part to the higher initial *Campylobacter* load, the citric acid may once again be ineffective at lowering *Campylobacter* load on the bird carcasses. The switch in the primary chiller from chlorine to PAA + caustic NaOH reduced *Campylobacter* loads on the 2nd Monday bird samples to uncountable levels, whereas countable *Campylobacter* loads were observed when chlorine was used during the 1st Monday sampling, suggesting PAA as a stronger antimicrobial. The removal of the spray bars did not affect the number of *Campylobacter*-positive carcass samples, and with the strong reduction of *Campylobacter* in the primary chiller with the use of PAA, a new post-chill intervention may not be required.

Plant F. The reduction of *Campylobacter* during all sampling periods can be seen at each intervention point along the production line (Fig 6.1-6.2). Comparing the Monday and Thursday bird samples of the 1st sampling period showed significantly ($p<0.05$) lower *Campylobacter* counts in the Thursday bird sampling period after the IOBW and lower *Campylobacter* counts along the production line, however there were more *Campylobacter*-positive bird samples (Table 6.2) after exiting the primary chiller and after exiting the dip tank, indicating some potential cross-contamination or microbial buildup in the immersion tanks. When the 2nd Monday and Thursday sampling periods were compared the significantly ($p<0.05$) lower *Campylobacter* counts post-pick may indicate that there is no cross-contamination during the 2nd sampling period. The initial *Campylobacter* load from the Thursday manure sample was also lower and there was a slight increase in *Campylobacter* counts after the pre-chiller, as well as

increased *Campylobacter*-positive samples after exiting the primary chiller and after exiting the dip tank, and so microbial buildup may be occurring in the immersion tanks.

Comparing the bird samples of the two Monday sampling periods and the two Thursday sampling periods no significant differences in *Campylobacter* counts were observed between the 1st and 2nd Monday sampling periods, and is to be expected as no in-plant changes were made between the 1st and 2nd sampling sets. Significantly ($p<0.05$) higher *Campylobacter* counts exiting the IOBW were observed in the 2nd Thursday bird samples, however higher post-pick levels were observed as well and this variation may be due to the new scalder removing slightly less organic matter than the old scalder. In the Monday sampling set and Thursday sampling set the number of *Campylobacter*-positive and *Salmonella*-positive bird samples did not change much which was also expected (Tables 6.1-6.2), however *Salmonella*-positive samples were taken from the scalder tank in the 2nd Thursday sampling period, which may again be due to the new scalder not reducing microbial load as much as the old scalder.

5. Overall Conclusions

Several plants displayed a microbial build-up and higher survival rate of pathogens when the Thursday samples were compared to the Monday samples. Proper cleaning of equipment as well as a regular influx of fresh water and antimicrobials into the immersion tanks is crucial to the prevention higher contamination rates as a plant reaches the end of the production week. If proper cleaning and disinfecting does not occur, the effectiveness of intervention methods may be reduced, and performance standards may not be met later in the production week. Any

sampling and in-plant pathogen testing during the beginning of the production week may also be skewed as they are occurring before the buildup occurs and increases microbial load.

The variation of incoming bacterial load between plants and within each sampling period at a specific plant seems to be another factor that determines the level of microbial reduction as a result of the use of antimicrobials interventions in the processing plants. The incoming *Campylobacter* load from the farms can affect whether or not the antimicrobials and point of application of intervention are effective at reducing the pathogen to below detectable levels. One cause of this variation could have been the time of year each sampling period occurred. The first sampling period occurred during the winter months, whereas the second sampling period occurred during the summer months. The rise of temperatures in the summer months leads to heat stress in broilers, increased fecal shedding, higher pathogen survival, and easier contamination. While the summer sampling did not increase the incoming bacterial load at all plants, it is something that should be considered when determining the proper in-plant intervention methods to use. That being a factor, there still seems to be noticeable differences in microbial reduction between intervention methods.

The use of PAA was observed to be the most effective antimicrobial for use within the primary chiller and dip tanks of processing plants, with lower log CFU/mL counts as well as fewer *Campylobacter*-positive bird samples than the plants that used chlorine. In addition, the use of Cetylpyridinium Chloride (CPC) in a spray cabinet as a post dip tank intervention point displayed the ability to further reduce *Campylobacter* counts to below detectable levels when the use of a dip tank was not sufficient. The use of chemicals in the scald tanks does not seem to be effective at lowering the microbial load on bird samples. The use of a post-chill

intervention displayed further reduction of microbial load in almost all samples, and should be considered as a strong method to further reduce pathogen counts on poultry. The use of a 2nd pre-chiller also displayed a microbial reduction on bird samples entering the primary chiller; however this may be inefficient if the primary chiller and post-chill intervention are sufficient to reduce pathogens to below-detectable levels. Slight levels of microbial buildup and cross-contamination were displayed in the immersion tanks of the processing plants, and the proper cleaning and maintenance of these immersion tanks is paramount to keeping this risk low.

6. Future Research Potential

It has been shown that post-chill interventions can further reduce bacterial load on poultry carcasses, however the comparison between the different methods has not been researched. Comparing the reduction of a finishing chiller versus a post-chill dip tank and a post chill drip would be beneficial so that the optimal post-chill intervention could be implemented in the plants. Additionally, testing PAA levels in a finishing chiller with a sensory panel should be conducted so that the highest parts per million (ppm) concentration of PAA with minimized quality loss can be achieved. *Campylobacter* is also known to enter a viable but non-culturable (VBNC) state when stressed, and so even with negative enrichments, further testing to confirm cell death and not VBNC state after the post-chill intervention may be necessary. Follow-up research on the microbial reduction effects of CPC use at various stages in a processing plant would be beneficial to determine the optimum placement for a CPC spray cabinet along the production line.

References

1. Acheson, D., and B. M. Allos. 2001. *Campylobacter jejuni* infections: update on emerging issues and trends. *Clin Infect Dis.* 32:1201-1206.
2. Allos, B. M. 1997. Association between *Campylobacter jejuni* infection and Guillain-Barré syndrome. *J Infect Dis.* (Suppl 2):S125-128.
3. Anonymous. 1998. Benefits and limitations of antimicrobial treatments for poultry carcasses. Scientific Committee on Veterinary Measures Relating to Public Health. Available at: http://europa.eu.int/comm/food/fs/sc/scv/out14_en.html. Accessed 23 August 2013
4. Anonymous. 2000. CDC *Salmonella* surveillance summaries 1976-1999. Centers for Disease Control and Prevention. Public Health Laboratory Information System. Washington: U.S. Government printing Office.
5. Anonymous. 2011. CDC estimates of foodborne illness in the United States. Centers for Disease Control and Prevention. Available at: http://www.cdc.gov/foodborneburden/2011-foodborne-estimates.html. Accessed 23 August 2013.
6. Anonymous. 2011. Incidence and trends of foodborne illness, 2011. Centers for Disease Control and Prevention. Available at: http://www.cdc.gov/features/dsfoodnet/. Accessed 23 August 2013.
7. Anonymous. 2012. Performance standards for *Salmonella* and *Campylobacter* in chilled carcasses at young chicken and turkey slaughter establishments. United States Department of Agriculture. Food Safety and Inspection Service. Washington, D. C. Available at: http://www.fsis.usda.gov/OPPDE/rdad/FSISNotices/54-12.pdf Accessed 23 August 2013.
8. Anonymous. 2013. Safe and suitable ingredients used in the production of meat, poultry, and egg products. United States Department of Agriculture. Food Safety and Inspection Service. Washington, D. C. Available at: http://www.fsis.usda.gov/OPPDE/rdad/FSISDirectives/7120.1.pdf. Accessed 23 August 2013.
9. Arritt, F. M., Eifert, J. D., Pierson, M. D., and S. S. Sumner. 2002. Efficacy of antimicrobials against *Campylobacter jejuni* on chicken breast skin. *J Appl Poult Res.* 11:358-366.

10. Avila, L. A. F., Nascimento, V. P., Salle, C. T. P., and H. L. S. Moraes. 2006. Effects of probiotics and maternal vaccination in *Salmonella* Enteritidis infection in broiler chicks. *Avian Dis Dig*. 1:e22-e22.
11. Bailey, J. S., Stern, N. J., Fedorka-Cray, P., Craven, S. E., Cox, N. A., Cosby, D. E., Ladely, S., and M. T. Musgrove. 2001. Sources and movement of *Salmonella* through integrated poultry operations: a multistate epidemiological investigation. *J Food Prot*. 64:1690-1697.
12. Bashor, M. P., Curtis, P. A., Keener, K. M., Sheldon, B. W., Kathariou, S., and J. A. Osborne. 2004. Effects of carcass washers on *Campylobacter* contamination in large broiler processing plants. *Poult Sci.* 83:1232-1239.
13. Batz, M. B., Hoffman, S. and J. G. Morris. 2011. Ranking the risks: the 10 pathogen-food combinations with the greatest burden on public health. Emerging Pathogens Institute. Available at: http://www.epi.ufl.edu/sites/www.epi.ufl.edu/files/RankingTheRisksREPORT.pdf. Accessed 23 August 2013.
14. Bauermeister, L. J., Bowers, J. W., Townsend, J. C., 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.
15. Beers, K., Rheingans, J., Chinault, K., Cook, P., Smith, B., and A. Waldroup. 2006. Microbial efficacy of commercial application of Cecure® CPC antimicrobial to ingesta-contaminated pre-chill broiler carcasses. *Int J Poult Sci.* 5:698-703.
16. 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.* 9:43-47
17. Berrang, M. E., Windham, W. R., 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.* 90:896-900.
18. Blaser, M. J., Smith, P. F., Wang, W. L., and J. C. Hoff. 1986. Inactivation of *Campylobacter jejuni* by chlorine and monochloramine. *Appl Environ Microbiol*. 51:307-311.
19. Brake, J., and B. W. Sheldon. 1990. Effect of a quaternary ammonium sanitizer for hatching eggs on their contamination, permeability, water loss, and hatchability. *Poult Sci.* 69:517-525.
20. Cabrita, J., Rhodrigues, J., Braganca, F., Morgado, C., Pires, I., and A. P. Goncalves. 1992. Prevalence, biotypes, plasmid profile, and antimicrobial resistance of *Campylobacter* isolated from wild and domestic animals from northeast Portugal. *J Appl Bacteriol.* 73:279-285.

21. Carramiñana, J. J., Rota, C., Agustín, I., and A. Herrera. 2004. High prevalence of multiple resistance to antibiotics in *Salmonella* serovars isolated from a poultry slaughterhouse in Spain. *Vet Microbiol.* 104:133-139.
22. Cason, J. A., and A. Hinton Jr. 2006. Coliforms, *Escherichia coli, Campylobacter,* and *Salmonella* in a counterflow poultry scalder with a dip tank. *Int J Poult Sci*. 5:846-849.
23. Chantarapanont, W. Berrang, M. E., 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*. 67:1146-1152.
24. Davies, R. H., and M. Breslin. 2003. Observations on *Salmonella* contamination of commercial laying farms before and after cleaning and disinfection. *Veterinary Record*. 152:283-287.
25. Davies, R. H., and C. Wray. 1995. Contribution of the lesser mealworm beetle (*Alphitobius diaperinus*) to carriage of *Salmonella* Enteritidis in poultry. *Vet Rec.* 137:407-408.
26. Friedman, C. R., Neimann, J., Wegener, H.C., and R. V. Tauxe. 2000. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. In: Nachamkin, I., and M. J. Blaser, eds. *Campylobacter.* 2nd Edition. Washington, DC: ASM Press. 121-138.
27. Heyndrickx, M., Vandekerchove, D., Herman, L., Rollier, I., Grijspeerdt, K., and L. De Zutter. 2002. Routes for *Salmonella* contamination of poultry meat: epidemiological study from hatchery to slaughterhouse. *Epidemiol Infect.* 129:253-265.
28. Humphrey, T. J., Henley, A., and D. G. Lanning. 1993. The colonization of broiler chickens with *Campylobacter jejuni*: some epidemiological investigations. *Epidemiol Infect.* 110:601-607.
29. Hwang, C. A., and L. R. Beuchat. 1995. Efficacy of selected chemicals for killing pathogenic and spoilage microorganisms on chicken skin. *J Food Prot.* 58:19-23.
30. Jacobs-Reitsma, W., and N. Bolder. 1998. The role of transportation crates in *Campylobacter* contamination of broilers. Porc. 9th Int. Workshop on *Campylobacter, Helicobacter,* and related organisms. Lastovica, A. J., Newell, D. G., and E. E. Lastovica, ed. Capetown South Africa. 379-380.
31. Jacobs-Reitsma, W. F., van de Giessen, A. W., Bolder, N. M., and R. W. A. W. Mulder. 1995. Epidemiology of *Campylobacter* spp. at two Dutch broiler farms. *Epidemiol Infect.* 114:413-421
32. James, W. O., Brewer, R. L., Prucha, J. C., Williams, W. O., and D. R. Parham. 1992. Effects of chlorination of chill water on the bacteriologic profile of raw chicken carcasses and giblets. *J Am Vet Med Assoc*. 200:60-63.
33. Kameyama, M., Chuma, T., Nishimoto, T., Oniki, H., Yanagitani, Y., Kanetou, R., Gotou, K., Shahada, F., Iwata, H., and K. Okamoto. 2012. Effect of cooled and chlorinated chiller

water on *Campylobacter* and coliform counts on broiler carcasses during chilling at a middle-size poultry processing plant. *J Vet Med Sci.* 74:129-133.

34. Kazwala, R. R., Collins, J. D., Hannan, J., Crinion, R. A., and H. O'Mahony. 1990. Factors responsible for the introduction and spread of *Campylobacter jejuni* infection in commercial poultry production. *Vet Rec.* 126:305-306.
35. Keener, K. M., Bashor, M. P., Curtis, P. A., Sheldon, B. W., and S. Kathariou. Comprehensive review of *Campylobacter* and poultry processing. Comprehensive Reviews in Food Science and Food Safety. 3:105-116.
36. Kemp, G. K., Aldrich, M. L., Guerra, M. L., and K. R. Schneider. 2001a. Continuous online processing of fecal- and ingesta-contaminated poultry carcasses using acidified sodium chlorite antimicrobial intervention. *J Food Prot.* 64:807-816.
37. Kemp, G. K., Aldrich, M. L., and A. L. Waldroup. 2001b. Acidified sodium chlorite antimicrobial treatment of broiler carcasses. *J Food Prot.* 63:1087-1092.
38. King, E. O. 1957. Human infections with *Vibrio fetus* and a closely related *Vibrio*. *J Infect Dis.* 101:119-128.
39. Li, Y., Slavik, M. F., Walker, J.T., and H. Xiong. 1997. Pre-chill spray of chicken carcasses to reduce *Salmonella* Typhimurium. *J Food Sci.* 62:605-607.
40. Marin, C., Balasch, S., Vega, S., and M. Lainez. 2011. Sources of *Salmonella* contamination during broiler production in Eastern Spain. *Prev Vet Med.* 98:39-45.
41. McKee, S. R., Townsend, J. C., and S. F. Bilgili. 2008. Use of a scald additive to reduce levels of *Salmonella typhimurium* during poultry processing. *Poult Sci.* 87:1672-1677.
42. Mead, G. C., Adams, B. W., and R. T. Parry. 1975. The effectiveness of in-plant chlorination in poultry processing. *Br Poult Sci.* 16:517-526.
43. Mishu, B., Koehler, J., Lee, L., Rodrique, D., Brenner, F., Blake, P., and R. V. Tauxe. 1994. Outbreaks of *Salmonella* Enteritidis infections in the United States, 1985-1991. *J Infect Dis.* 169:547-552.
44. Montrose, M. S., Shane, S. M., and K. S. Harrington. 1985. Role of litter in the transmission of *Campylobacter jejuni. Avian Dis.* 29:392-399.
45. Nagel, G. M., Bauermeister, L. J., Bratcher, C. L., Singh, M., 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.* 165:281-286.
46. Newell, D. G. and J. A. Wagenaar. 2000. Poultry infections and their control at the farm level. In: *Campylobacter*, 2nd Edition. Nachamkin, I., and M. J. Blaser (eds.) Washington, DC: American Society for Microbiology. 497-509.
47. Northcutt, J. K., and D. R. Jones. 2004. A survey of water use and common industry practices in commercial broiler processing facilities. *J Appl Poult Res*. 13:48-54.

48. Northcutt, J. K., Smith, D. P., Musgrove, M. T., Ingram, K. D., and A. Hinton Jr. 2005. Microbiological impact of spray washing broiler carcasses using different chlorine concentrations and water temperatures. *Poult Sci.* 84:1648-1652.
49. Olsen, A. R., and T. S. Hammack. 2000. Isolation of *Salmonella* spp. from the housefly, *Musca domestica* L., and the dump fly, *Hydrotaea aenescens* (Wiedemann) (Diptera: Muscidae), at Caged-Layer Houses. *J Food Prot*. 63:958-960.
50. Olsen, J. E., Brown, D. J., Madsen, M., and M. Bisgaard. 2003. Cross-contamination with *Salmonella* on a broiler slaughterhouse line demonstrated by use of epidemiological markers. *J Appl Microbiol.* 94:826-835.
51. 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.* 68:1752-1760.
52. Oyarzabal, O. A., Hawk, C., Bilgili, S. F., Warf, C. C., and G. K. Kemp. 2004. Effects of postchill application of acidified sodium chlorite to control *Campylobacter* spp. and *Escherichia coli* on commercial broiler carcasses.
53. Patrick, M. E., Adcock, P. M., Gomes, T. M., Altekruse, S. F., Holland, B. H., Tauxe, R. V., and D. L. Swerdlow. 2004. *Salmonella* Enteritidis infections, United States, 1985-1999. *Emerg Infect Diseases.* Vol 10.
54. Patterson, M. F. 1995. Sensitivity of *Campylobacter* spp. to irradiation in poultry meat. *Lett Appl Microbiol.* 20:338-340.
55. Pearson, A. D., Greenwood, M. H., Feltham, R. K., Healking, T. D., Donaldson, J., Jones, D. M., and R. R. Colwell. 1996. Microbial ecology of *Campylobacter jejuni* in a United Kingdon chicken supply chain: intermittent common source, vertical transmission, and amplification by flock propagation.
56. Pearson, A. D., Greenwood, M. H., Healing, T. D., Rollins, D., Shahamat, M., Donaldson, J., and R. R. Colwell. 1993. Colonization of broiler chickens by waterborne *Campylobacter jejuni. Appl Environ Microbiol.* 59:987-996.
57. Peterson, M. C. 1994. Clinical aspects of *Campylobacter jejuni* infections in adults. *West J Med.* 161:148-152.
58. L. J. V. Piddock. 1995. Quinolone resistance and *Campylobacter* spp. *J Antimicrob Chemother.* 36:891-898.
59. Poppe, C., Barnum, D. A., and W. R. Mitchell. 1986. Effect of chlorination of drinking water on experimental *Salmonella* infection in poultry. *Avian Dis.* 30:362-369.
60. Rice, B. E., Rollins, D. M., Mallinson, E. T., Carr, L., and S. W. Joseph. 1997. *Campylobacter jejuni* in broiler chickens: colonization and humoral immunity following oral vaccination and experimental infection. *Vaccine*. 15:1922-1932.
61. Robinson, D. A. 1981. Infective dose of *Campylobacter jejuni* in milk. *Br Med J.* 282:1584

62. Rodriguez de Ledesma, A. M., Riemann, H. P., and T. B. Farver. 1996. Short-time treatment with alkali and/or hot water to remove common pathogenic and spoilage bacteria from chicken wing skin. *J Food Prot.* 59:746-750.
63. Rose, N., Beaudeau, F., Drouin, P., Toux, J. Y., Rose, V., and P. Colin. 2000. Risk factors for *Salmonella* persistence after cleansing and disinfection in French broiler-chicken houses. *Prev Vet Med.*44:9-20.
64. Rosef, O., and G Kapperud. 1983. House flies (*Musca domestica*) as possible vectors of *Campylobacter fetus* subsp. jejuni. *Appl Environ Microbiol*. 45:381-383
65. S. M. Russell. 2008. The effect of an acidic, copper sulfate-based commercial sanitizer on indicator, pathogenic, and spoilage bacteria associated with broiler chicken carcasses when applied at various intervention points during poultry processing. *Poult Sci.* 87:1435-1440.
66. Sánchez, M. X., Fluckey, W. M., Brashears, M. M., 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.* 65:948-956.
67. Shreeve, J. E., Toszeghy, M., Pattison, M., and D. G. Newell. 2000. Sequential spread of *Campylobacter* infection in a multi-pen broiler house. *Avian Dis.* 46:378-385.
68. Slader, J., Domingue, G., Jørgensen, F., McAlpine, K., Owen, R. J., Bolton, F. J., and T. J. Humphrey. 2002. Impact of transport crate reuse and of catching and processing on *Campylobacter* and *Salmonella* contamination of broiler chickens. *Appl Environ Microbiol*. 68:713-719.
69. Slavik, M. F., Jeong-Weon, K., and J. T. Walker. 1995. Reduction of *Salmonella* and *Campylobacter* on chicken carcasses by changing scalding temperature. *J Food Prot*. 58:689-691
70. Smigic, N., Rajkovic, A., Arneborg, N., Siegumfeldt, H., Devlieghere, F., and D. S. Nielsen. 2011. Intracellular pH in *Campylobacter jejuni* when treated with aqueous chlorine dioxide. *Foodborne Pathog Dis.* 8:325.
71. Smith, T. 1894. The hog-cholera group of bacteria. *US Bur Anim Ind Bull* 6:6-40
72. Somers, E. B., Schoeni, J. L., and A. C. Wong. 1994. Effect of trisodium phosphate on biofilm and planktonic cells of *Campylobacter jejuni, Escherichia coli* O157: H7, *Listeria monocytogenes*, and *Salmonella* Typhimurium. *Int J Food Microbiol.* 22:269-276.
73. Stern, N. J., and J. E. Line. 1992. Comparison of three methods for recovery of *Campylobacter* spp. from broiler carcasses. *J Food Prot.*55:663-666.
74. Stern, N. J., Robach, M. C., Cox, D. F., and M. T. Musgrove. 2002. Effect of drinking water chlorination on *Campylobacter* spp. colonization of broilers. *Avian Dis*. 46:401-404.

75. Thiessen, G. P., Usborne, W. R., and H. L. Orr. 1984. The efficacy of chlorine dioxide in controlling *Salmonella* contamination and its effect on product quality of chicken broiler carcasses. *Poult Sci.* 63:647-653.
76. van de Giessen, A., Mazurier, S. I., Jacobs-Reitsma, W., Jansen, W., Berkers, P., Ritmeester, W., and K. Wernars. 1992. Study on the epidemiology and control of *Campylobacter jejuni* in poultry broiler flocks. *Appl Environ Microbiol.* 58:1913-1917.
77. Véron, M., and Chatelain, R. 1973. Taxonomic Study of the genus *Campylobacter* Sebald and Véron and designation of the neotype strain for the type species, *Campylobacter fetus* (Smith and Taylor) Sebald and Véron. *Int J Syst Bacteriol.* 23:122-134.
78. Waldroup, A. L., Beers, K. L., Cook, P. E., Dell, E. A., Odglen, R., Baker, R. A., Coleman, C. W., Smith, B. A., and B. W. Maingi. 2010. The effects of cetylpiridinium chloride (Cecure® CPC antimicrobial) on *Campylobacter* spp. on raw poultry: a review. *Int J Poult Sci.* 4:305-308.
79. Waldroup, A. L., Rathgeber, B. M., Forsythe, R. H., 1992. Effects of six modifications on the incidence and levels of spoilage and pathogenic organisms on commercially processed postchill broilers. *J ppl Poultry Res.* 1:226-234.
80. Wempe, J. M., Genigeorgis, C. A., Farver, T. B., and H. I. Yusufu. 1983. Prevalence of *Campylobacter jejuni* in two California chicken processing plants. *Appl Environ Microbiol.* 45:355-359.
81. Whyte, P., Collins, J.D., McGill, K., Monahan, C., and H. O'Mahony. 2001. Quantitative investigation of the effects of chemical decontamination procedures on the microbiological status of broiler carcasses during processing. *J Food Prot*. 64:179-183.
82. Yang, H., Li, Y., and M. G. Johnson. 2001. Survival and death of *Salmonella* Typhimurium and *Campylobacter jejuni* in processing water and on chicken skin during poultry scalding and chilling. *J Food Prot.* 64:770-776.

Tables

Plant A *Salmonella* Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	-	-	+	+
Scalder Water 1	+	+	+	-
Scalder Water 2	+	+	-	-
Scalder Water 3	-	-	-	-
Pre-Chiller Water	-	+	-	-
Pre-Chiller #2	-	-	-	-
Primary Chiller Water	-	-	-	-
Bird Samples				
Post Pick	0/5	0/5	0/5	0/5
Pre-Chill	0/5	0/5	0/5	0/5
Pre-Post	0/5	0/5	0/5	0/5
Post Chill	0/5	1/5	0/5	0/5
Post Dip	0/5	0/5	0/5	0/5

Table 1.1 Prevalance of *Salmonella* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), 3rd scalder tank (Scalder Water 3), 1st pre-chiller (Pre-Chiller Water), 2nd pre-chiller (Pre-Chiller #2) and primary chiller (Primary Chiller Water). **Bird Samples:** Number of *Salmonella*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill), and after exiting the post-chill intervention (Post Dip).

Plant A *Campylobacter* Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	+	+	+	+
Scalder Water 1	+	+	+	+
Scalder Water 2	+	-	+	+
Scalder Water 3	+	+	+	+
Pre-Chiller Water	+	+	+	+
Pre-Chiller #2	+	-	-	-
Primary Chiller Water	-	+	-	-
Bird Samples				
Post Pick	5/5	5/5	5/5	5/5
Pre-Chill	5/5	5/5	5/5	5/5
Pre-Post	1/5	2/5	5/5	5/5
Post Chill	1/5	1/5	1/5	2/5
Post Dip	0/5	0/5	0/5	0/5

Table 1.2 Prevalance of *Campylobacter* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), 3rd scalder tank (Scalder Water 3), 1st pre-chiller (Pre-Chiller Water), 2nd pre-chiller (Pre-Chiller #2) and primary chiller (Primary Chiller Water). **Bird Samples:** Number of *Campylobacter*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill), and after exiting the post-chill intervention (Post Dip).

Plant B *Salmonella* Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	+	+	-	-
Scalder Water 1	+	+	-	+
Scalder Water 2	+	-	+	+
Scalder Water 3	+	+	+	-
Pre-Chiller Water	+	-	+	-
Primary Chiller Water	-	+	-	-
Dip Tank Water	n/a	n/a	-	-
Bird Samples				
Post Pick	3/5	0/5	2/5	0/5
Pre-Chill	3/5	0/5	1/5	4/5
Pre-Post	4/5	0/5	0/5	1/5
Post Chill	0/5	0/5	0/5	0/5
Post Dip	n/a	n/a	0/5	0/5

Table 2.1 Prevalance of *Salmonella* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), 3rd scalder tank (Scalder Water 3), pre-chiller (Pre-Chiller Water), primary chiller (Primary Chiller Water), and post chill dip tank (Dip Tank Water). **Bird Samples:** Number of *Salmonella*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill), and after exiting the post-chill intervention (Post Dip).

Plant B Campylobacter Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	+	+	+	+
Scalder Water 1	+	+	+	+
Scalder Water 2	+	+	+	+
Scalder Water 3	-	+	+	+
Pre-Chiller Water	+	+	+	+
Primary Chiller Water	-	-	+	-
Dip Tank Water	n/a	n/a	-	-
Bird Samples				
Post Pick	4/5	4/5	5/5	5/5
Pre-Chill	2/5	4/5	5/5	5/5
Pre-Post	0/5	5/5	5/5	5/5
Post Chill	0/5	2/5	2/5	3/5
Post Dip	n/a	n/a	0/5	2/5

Table 2.2 Prevalance of *Campylobacter* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), 3rd scalder tank (Scalder Water 3), pre-chiller (Pre-Chiller Water), primary chiller (Primary Chiller Water), and post chill dip tank (Dip Tank Water). **Bird Samples:** Number of *Campylobacter*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill), and after exiting the post-chill intervention (Post Dip).

Plant C *Salmonella* Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	-	-	-	-
Scalder Water 1	-	-	-	-
Scalder Water 2	-	-	-	-
Pre-Chiller Water	-	-	-	-
Primary Chiller Water	-	-	-	-
Dip Tank Water	-	-	-	+
Bird Samples				
Post Pick	0/5	0/5	0/5	0/5
Pre-Chill	0/5	0/5	0/5	1/5
Pre-Post	0/5	0/5	0/5	1/5
Post Chill	0/5	0/5	0/5	0/5
Post Dip	0/5	0/5	0/3	0/3
Post Cabinet	n/a	n/a	0/3	0/3

Table 3.1 Prevalance of *Salmonella* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), pre-chiller (Pre-Chiller Water), primary chiller (Primary Chiller Water), and post chill dip tank (Dip Tank Water). **Bird Samples:** Number of *Salmonella*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill), after exiting the post-chill dip tank (Post Dip), and after exiting the CPC spray cabinet (Post Cabinet).

Plant C *Campylobacter* Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	+	+	+	+
Scalder Water 1	+	+	+	+
Scalder Water 2	+	+	+	+
Pre-Chiller Water	+	-	+	+
Primary Chiller Water	-	-	+	+
Dip Tank Water	-	-	+	+
Bird Samples				
Post Pick	5/5	5/5	5/5	5/5
Pre-Chill	5/5	5/5	5/5	5/5
Pre-Post	5/5	5/5	5/5	5/5
Post Chill	5/5	4/5	1/5	5/5
Post Dip	5/5	0/5	1/3	3/3
Post Cabinet	n/a	n/a	0/3	0/3

Table 3.2 Prevalance of *Campylobacter* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), pre-chiller (Pre-Chiller Water), primary chiller (Primary Chiller Water), and post chill dip tank (Dip Tank Water). **Bird Samples:** Number of *Campylobacter*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill), after exiting the post-chill dip tank (Post Dip), and after exiting the CPC spray cabinet (Post Cabinet).

Plant D *Salmonella* Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	-	+	-	-
Scalder Water 1	-	-	-	-
Scalder Water 2	-	-	-	-
Scalder Water 3	-	-	-	-
Pre-Chiller Water	-	-	-	-
Primary Chiller Water	-	-	-	-
Bird Samples				
Post Pick	0/5	0/5	0/5	1/5
Pre-Chill	0/5	0/5	0/5	0/5
Pre-Post	0/5	0/5	0/5	0/5
Post Chill	0/5	0/5	0/5	0/5

Table 4.1 Prevalance of *Salmonella* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), 3rd scalder tank (Scalder Water 3), pre-chiller (Pre-Chiller Water), and primary chiller (Primary Chiller Water). **Bird Samples:** Number of *Salmonella*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), and after exiting the primary chiller (Post Chill).

Plant D *Campylobacter* Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	+	+	+	+
Scalder Water 1	+	-	+	+
Scalder Water 2	+	-	+	+
Scalder Water 3	+	+	+	+
Pre-Chiller Water	+	+	+	+
Primary Chiller Water	-	+	-	-
Bird Samples				
Post Pick	5/5	5/5	5/5	5/5
Pre-Chill	5/5	5/5	5/5	5/5
Pre-Post	5/5	5/5	3/5	5/5
Post Chill	5/5	5/5	2/5	5/5

Table 4.2 Prevalance of *Campylobacter* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), 3rd scalder tank (Scalder Water 3), pre-chiller (Pre-Chiller Water), and primary chiller (Primary Chiller Water). **Bird Samples:** Number of *Campylobacter*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), and after exiting the primary chiller (Post Chill).

Plant E *Salmonella* Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	+	-	+	+
Scalder Water 1	-	+	+	+
Scalder Water 2	-	-	+	-
Scalder Water 3	-	-	-	-
Pre-Chiller Water	-	-	-	-
Primary Chiller Water	-	-	-	-
Bird Samples				
Post Pick	1/5	0/5	2/5	0/5
Pre-Chill	0/5	2/5	0/5	0/5
Pre-Post	0/5	1/5	0/5	0/5
Post Chill	0/5	0/5	0/5	0/5
Post Dip	0/5	0/5	n/a	n/a

Table 5.1 Prevalance of *Salmonella* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), 3rd scalder tank (Scalder Water 3), pre-chiller (Pre-Chiller Water), and primary chiller (Primary Chiller Water). **Bird Samples:** Number of *Salmonella*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill), and after exiting the post-chill intervention (Post Dip).

Plant E *Campylobacter* Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	+	+	+	+
Scalder Water 1	+	+	+	+
Scalder Water 2	+	-	+	+
Scalder Water 3	+	-	-	-
Pre-Chiller Water	+	-	-	-
Primary Chiller Water	-	-	-	-
Bird Samples				
Post Pick	5/5	5/5	5/5	5/5
Pre-Chill	5/5	0/5	5/5	5/5
Pre-Post	5/5	0/5	2/5	5/5
Post Chill	2/5	0/5	5/5	4/5
Post Dip	4/5	0/5	n/a	n/a

Table 5.2 Prevalance of *Campylobacter* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), 3rd scalder tank (Scalder Water 3), pre-chiller (Pre-Chiller Water), and primary chiller (Primary Chiller Water). **Bird Samples:** Number of *Campylobacter*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill), and after exiting the post-chill intervention (Post Dip).

Plant F *Salmonella* Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	+	-	+	+
Scalder Water 1	-	-	+	+
Scalder Water 2	-	-	-	+
Scalder Water 3	-	-	-	-
Pre-Chiller Water	-	-	-	-
Primary Chiller Water	-	-	-	-
Dip Tank Water	-	-	-	-
Bird Samples				
Post Pick	0/5	0/5	0/5	0/5
Pre-Chill	0/5	1/5	0/5	0/5
Pre-Post	0/5	0/5	0/5	0/5
Post Chill	0/5	0/5	0/5	0/5
Post Dip	0/5	0/5	0/5	0/5

Table 6.1 Prevalance of *Salmonella* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), 3rd scalder tank (Scalder Water 3), pre-chiller (Pre-Chiller Water), primary chiller (Primary Chiller Water), and post chill dip tank (Dip Tank Water). **Bird Samples:** Number of *Salmonella*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill), and after exiting the post-chill intervention (Post Dip).

Plant F *Campylobacter* Enrichments

	1st Sampling (Winter)		2nd Sampling (Summer)	
Sample	Monday	Thursday	Monday	Thursday
Manure Grab	+	+	+	+
Scalder Water 1	-	+	+	+
Scalder Water 2	-	+	+	+
Scalder Water 3	+	+	+	+
Pre-Chiller Water	+	+	+	+
Primary Chiller Water	-	+	-	+
Dip Tank Water	-	-	-	-
Bird Samples				
Post Pick	5/5	5/5	5/5	5/5
Pre-Chill	5/5	5/5	5/5	5/5
Pre-Post	5/5	5/5	5/5	5/5
Post Chill	0/5	1/5	1/5	4/5
Post Dip	0/5	4/5	0/5	4/5

Table 6.2 Prevalance of *Campylobacter* in a shackling belt manure sample (Manure Grab), water samples taken from the 1st scalder tank (Scalder Water 1), 2nd scalder tank (Scalder Water 2), 3rd scalder tank (Scalder Water 3), pre-chiller (Pre-Chiller Water), primary chiller (Primary Chiller Water) and post chill dip tank (Dip Tank Water). **Bird Samples:** Number of *Campylobacter*-positive bird samples taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill), and after exiting the post-chill intervention (Post Dip).

Figures

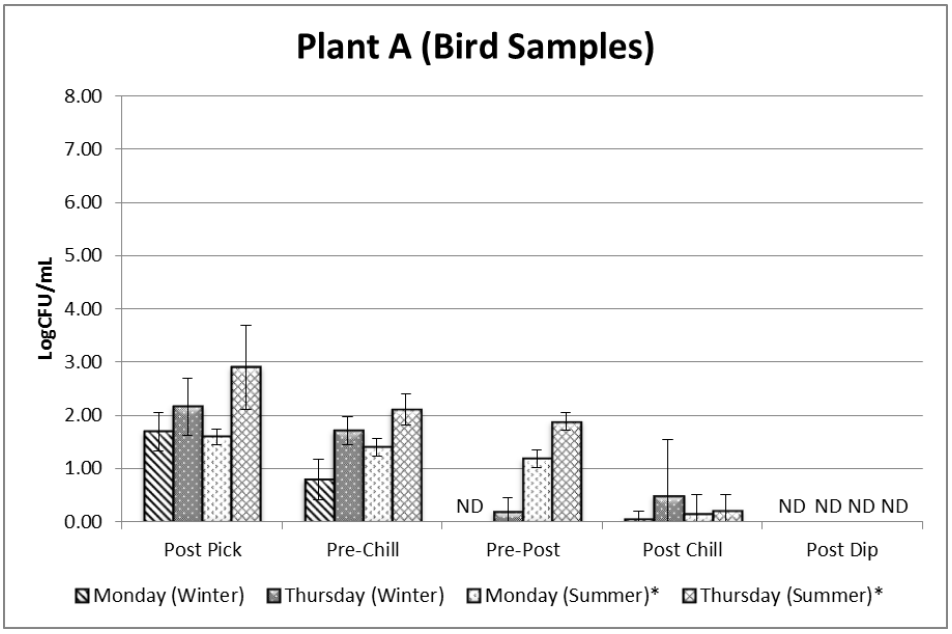

Fig. 1.1 Number (with standard deviation) of *Campylobacter spp.* recovered from bird carcass rinses taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill) and after exiting the post chill intervention (Post Dip). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)

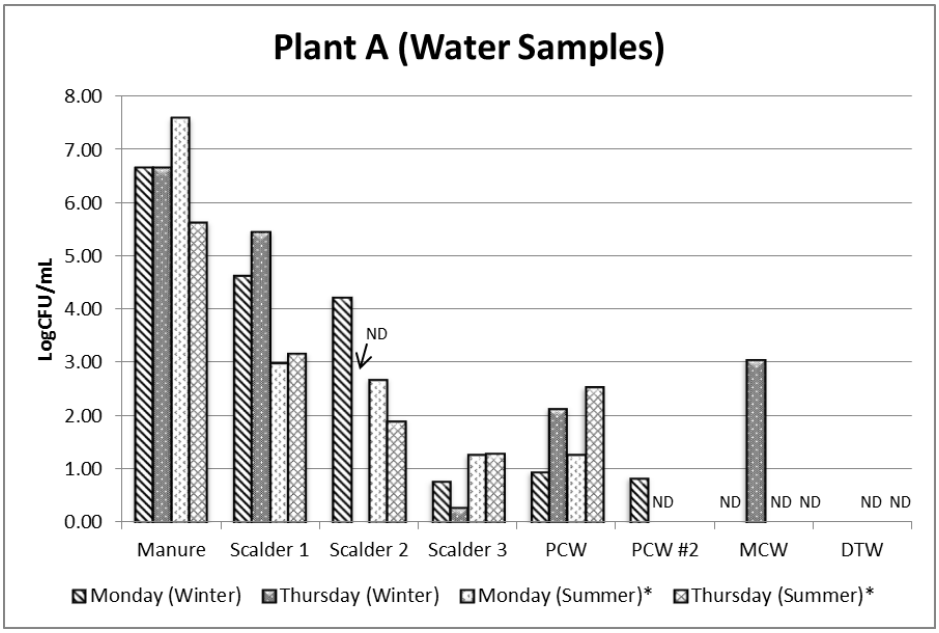

Fig. 1.2 Number of *Campylobacter spp.* recovered from a manure grab at the end of the shackling belt (Manure) and water samples taken from the 1st scalder tank (Scalder 1), 2nd scalder tank (Scalder 2), 3rd scalder tank (Scalder 3), 1st pre-chiller (PCW), 2nd pre-chiller (PCW #2), primary chiller (MCW), and from the post-chill dip tank (DTW). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)

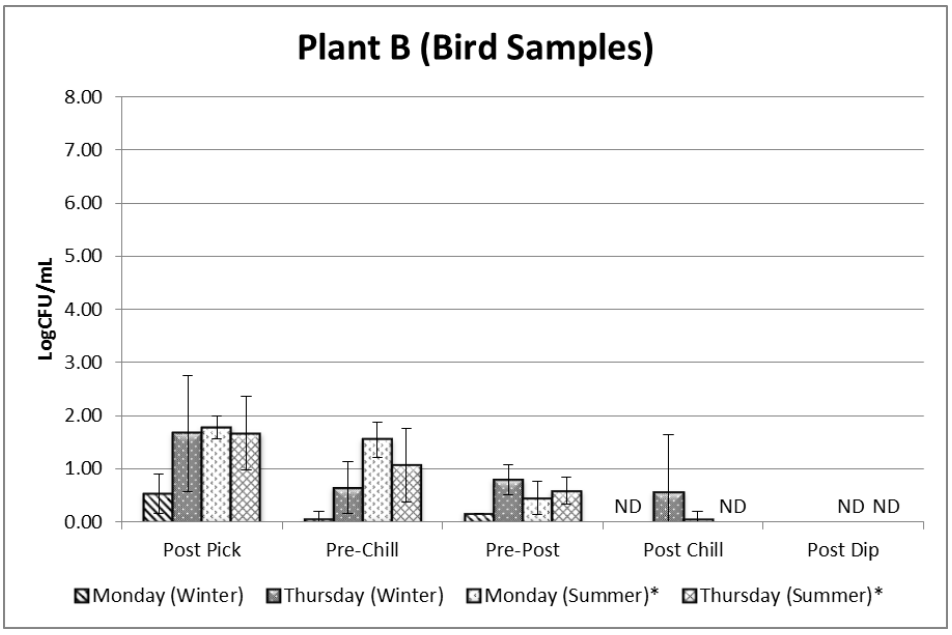

Fig. 2.1 Number (with standard deviation) of *Campylobacter spp.* recovered from bird carcass rinses taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill) and after exiting the post chill dip tank (Post Dip). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)

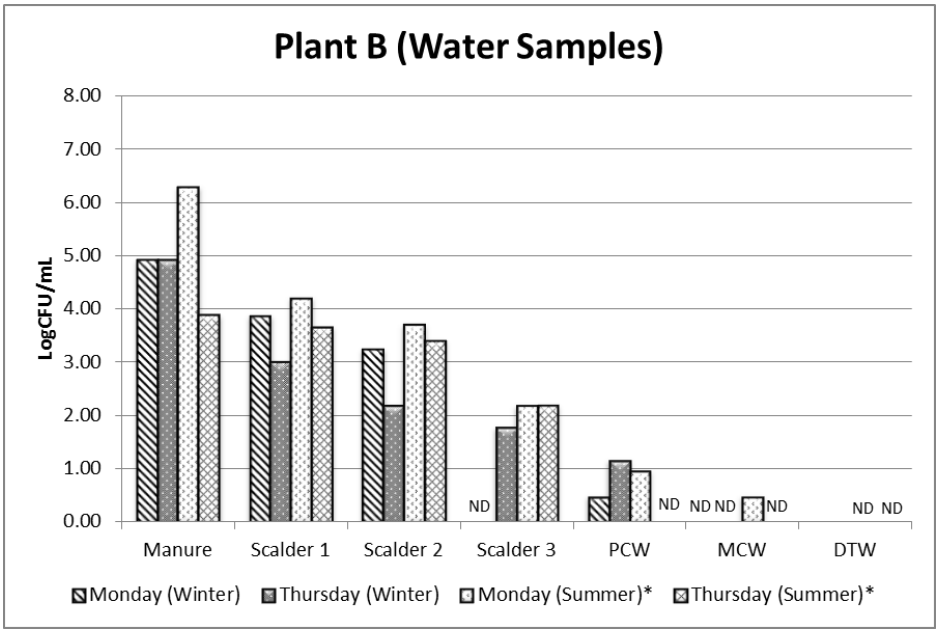

Fig. 2.2 Number of *Campylobacter spp.* recovered from a manure grab at the end of the shackling belt (Manure) and water samples taken from the 1st scalder tank (Scalder 1), 2nd scalder tank (Scalder 2), 3rd scalder tank (Scalder 3), pre-chiller (PCW), primary chiller (MCW), and from the post-chill dip tank (DTW). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)

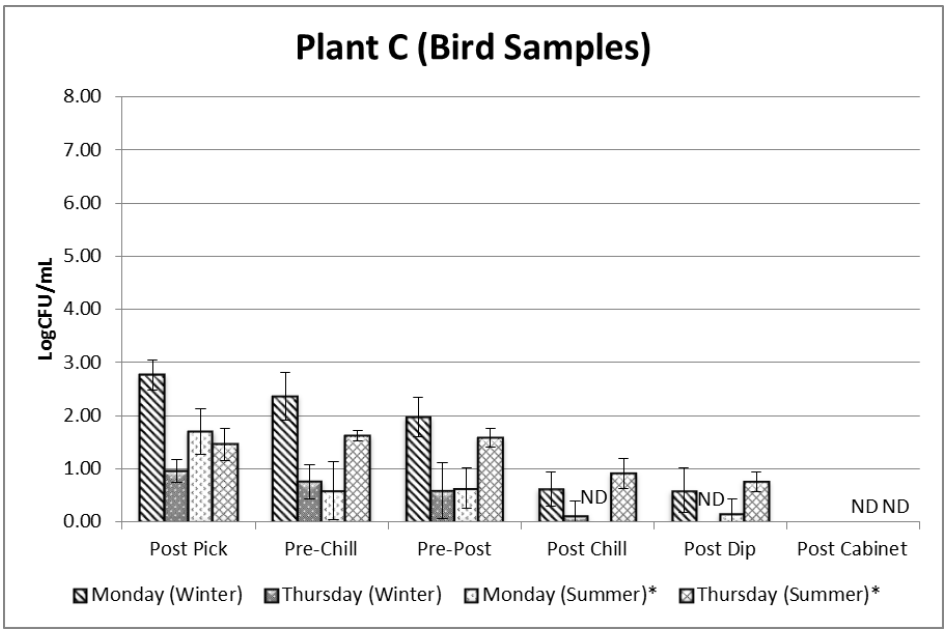

Fig. 3.1 Number (with standard deviation) of *Campylobacter spp.* recovered from bird carcass rinses taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill), after exiting the post chill dip tank (Post Dip), and after exiting the CPC spray cabinet (Post Cabinet). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)

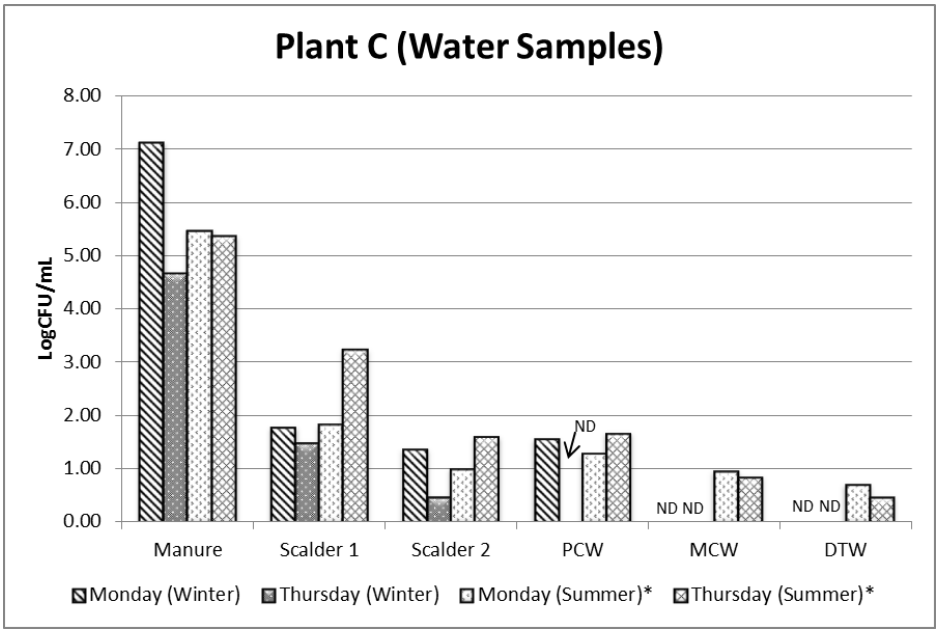

Fig. 3.2 Number of *Campylobacter spp.* recovered from a manure grab at the end of the shackling belt (Manure) and water samples taken from the 1st scalder tank (Scalder 1), 2nd scalder tank (Scalder 2), pre-chiller (PCW), primary chiller (MCW), and from the post-chill dip tank (DTW). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)

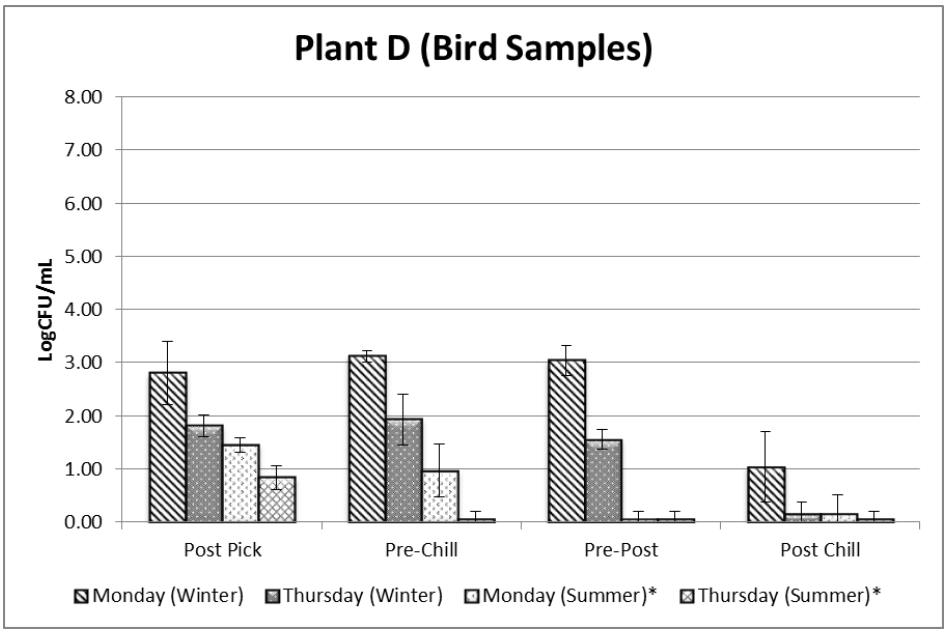

Fig. 4.1 Number (with standard deviation) of *Campylobacter spp.* recovered from bird carcass rinses taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), and after exiting the primary chiller (Post Chill). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)

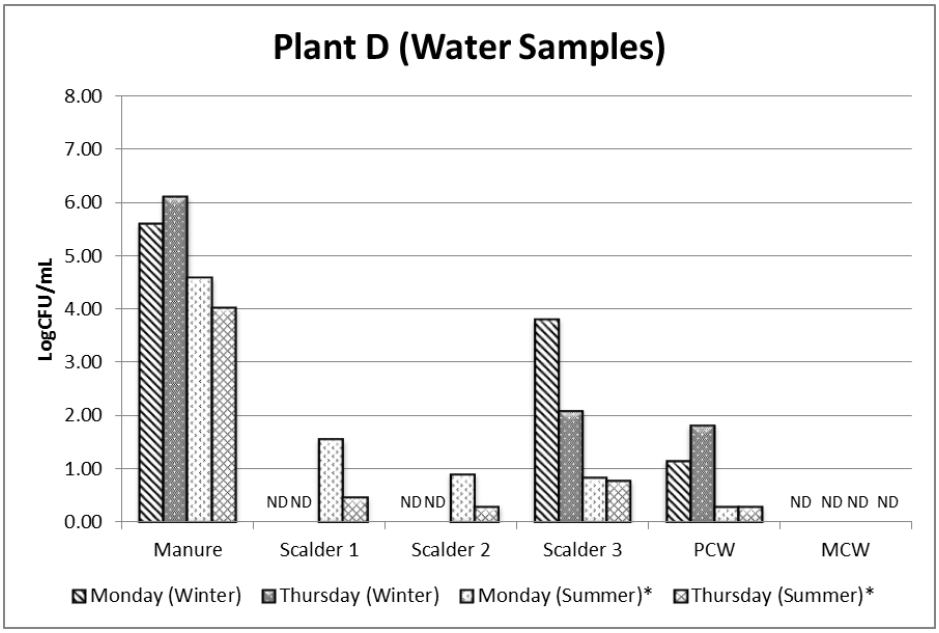

Fig. 4.2 Number of *Campylobacter spp.* recovered from a manure grab at the end of the shackling belt (Manure) and water samples taken from the 1st scalder tank (Scalder 1), 2nd scalder tank (Scalder 2), 3rd scalder tank (Scalder 3), pre-chiller (PCW), and from the primary chiller (MCW). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)

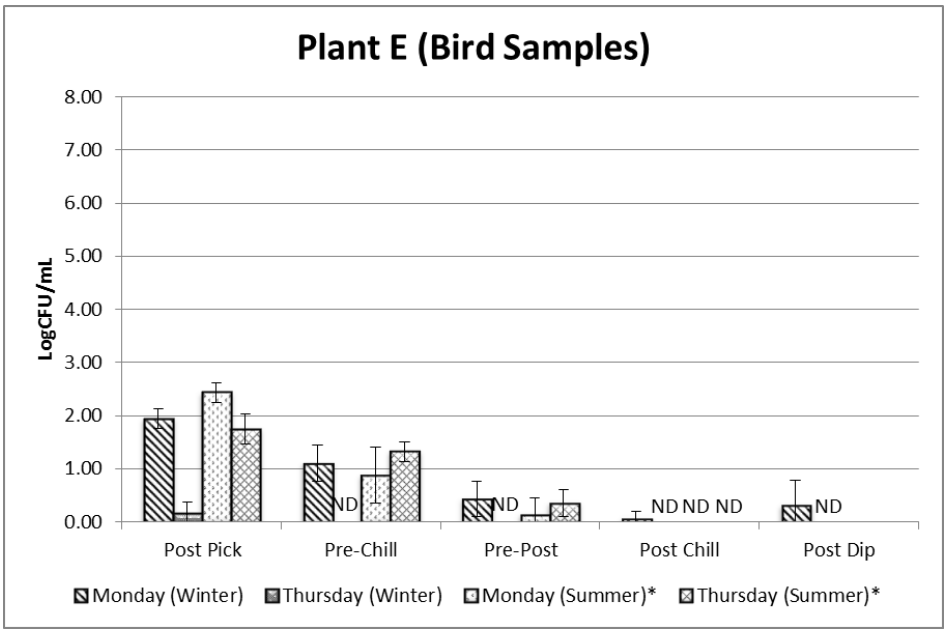

Fig. 5.1 Number (with standard deviation) of *Campylobacter spp.* recovered from bird carcass rinses taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill) and after exiting the post chill spray bars (Post Dip). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)

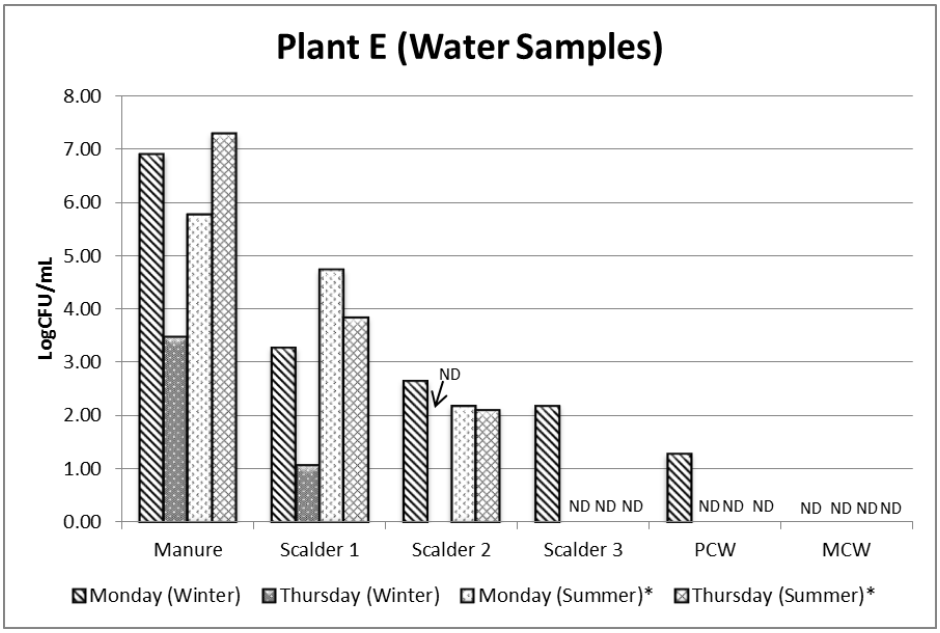

Fig. 5.2 Number of *Campylobacter spp.* recovered from a manure grab at the end of the shackling belt (Manure) and water samples taken from the 1st scalder tank (Scalder 1), 2nd scalder tank (Scalder 2), 3rd scalder tank (Scalder 3), pre-chiller (PCW), and from the primary chiller (MCW). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)

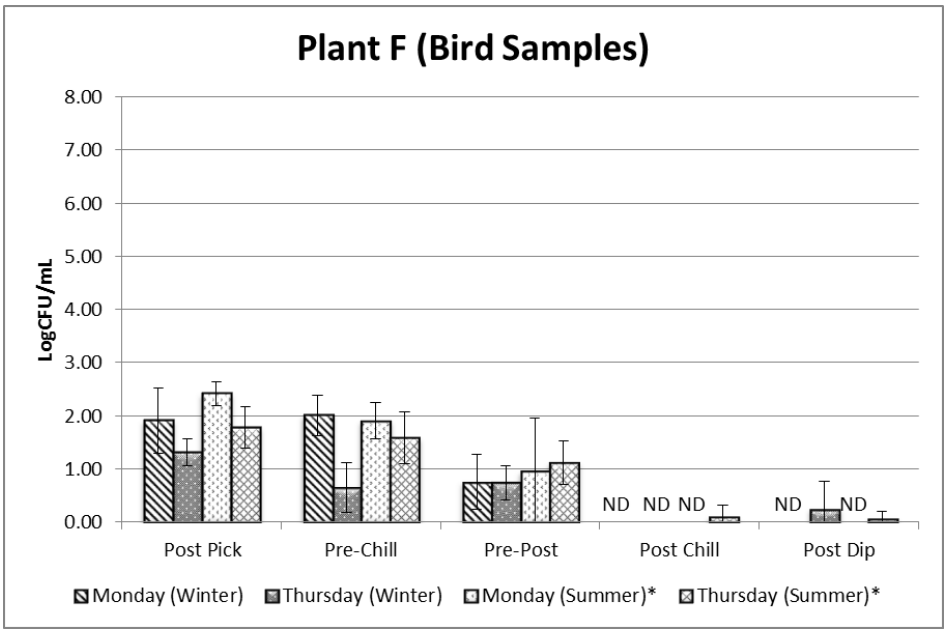

Fig. 6.1 Number (with standard deviation) of *Campylobacter spp.* recovered from bird carcass rinses taken before entering the IOBW (Post Pick), after exiting the IOBW (Pre-Chill), after exiting the pre-chiller (Pre-Post), after exiting the primary chiller (Post Chill) and after exiting the post chill dip tank (Post Dip). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)

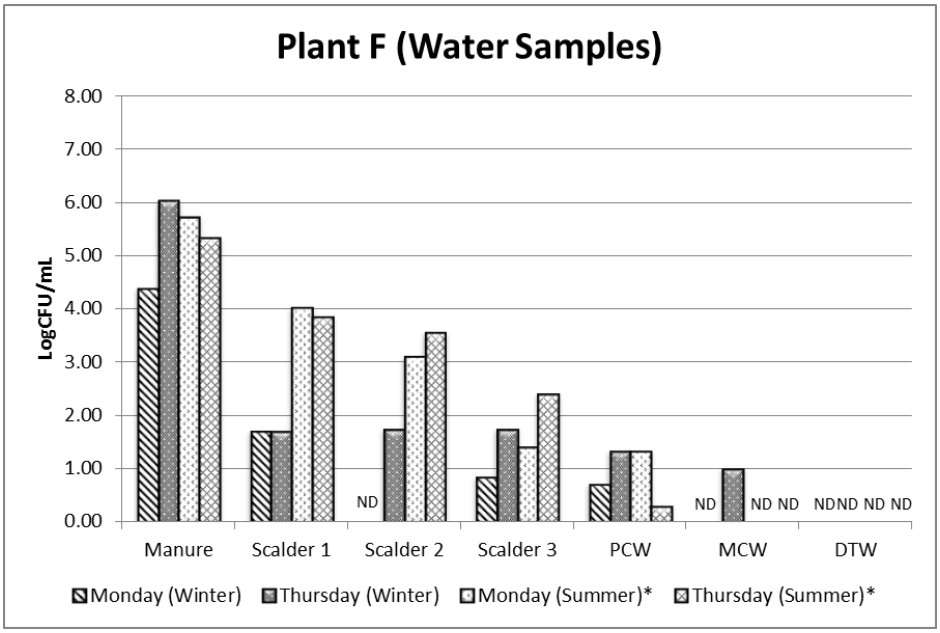

Fig. 6.2 Number of *Campylobacter spp.* recovered from a manure grab at the end of the shackling belt (Manure) and water samples taken from the 1st scalder tank (Scalder 1), 2nd scalder tank (Scalder 2), 3rd scalder tank (Scalder 3), pre-chiller (PCW), primary chiller (MCW), and from the post-chill dip tank (DTW). *Sampling taken after intervention changes were implemented. ND = none detected (Detection Limit = 1 CFU/mL)